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A Summary of Current Program, 7/1/63

and Preliminary Report of Progress

for 7/1/62 to 6/30/63

WESTERN UTILIZATION RESEARCH AND DEVELOPMENT DIVISION of the AGRICULTURAL RESEARCH SERVICE UNITED STATES DEPARTMENT OF AGRICULTURE

This progress report of U.S.D.A. and cooperative research is primarily a tool for use of scientists and administrators in program coordination, development and evaluation; and for use of advisory committees in program review and development of recommendations for future research programs.

The summaries of progress on U.S.D.A. and cooperative research include some tentative results that have not been tested sufficiently to justify general release. Such findings, when adequately confirmed will be released promptly through established channels. Because of this, the report is not intended for publication and should not be referred to in literature citations. Copies are distributed only to members of Department staff, advisory committee members and others having a special interest in the development of public agricultural research programs.

This report also includes a list of publications reporting results of U.S.D.A. and cooperative research issued between July 1, 1962, and June 30, 1963. Current agricultural research findings are also published in the monthly U.S.D.A. publication, Agricultural Research. This progress report was compiled in the Western Utilization Research and Development Division, Agricultural Research Service, U. S. Department of Agriculture, Albany, California.

UNITED STATES DEPARTMENT OF AGRICULTURE
Washington, D. C.
July 1, 1963

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INTRODUCTION

Utilization research on agricultural commodities deals with the discovery and development of new and improved products and the invention or perfection of processing technologies. The scientists, engineers, and technologists who carry on this research devote a substantial part of their concerted efforts to basic studies of physical and chemical properties of the commodities and products derived from them, in order to provide a firm base of exact knowledge for applied developments.

The present report summarizes the current research program of the Western Utilization Research and Development Division (one of four Utilization Divisions in the Agricultural Research Service), makes a report of progress toward the objectives of that program during Fiscal Year 1963, and describes a few of the more significant recent accomplishments of this work.

Research Area Covered by this Report

The farm commodities dealt with in this report are the cereal grains, wheat, rice, and barley; alfalfa and other forage crops; wool and mohair; citrus, apples, other fruits, and tree nuts; potatoes and other vegetables and dry beans and peas; castor and certain other oilseeds; sugar beets; new and replacement crops; and poultry and eggs.

Pharmacological research for all four of the Utilization Research Divisions is conducted at the Albany laboratory of the Western Division, and is described in this report.

Distinct phases of research on certain commodities having broad agricultural significance are pursued in other Utilization Research Divisions than the Western Division: Research on industrial uses of wheat and on milling technology is carried on in the Northern Division; certain areas of research on deciduous fruits and on potatoes and other vegetables are handled by the Eastern Division; particular lines of research on rice, vegetables, and fruits are carried on in the Southern Division. Research on new and replacement crops is carried on in all four Utilization Divisions.

Aims of Research on These Commodities

The group of commodities discussed here provides the nation with more than half of its food, either directly (cereal grains, fruits and vegetables, poultry meat and eggs, and beet sugar) or indirectly through feeding of meat animals (forage crops, wheat, barley). The other commodities in the group supply us with our most important animal fibers (wool and mohair) and offer opportunities of development into numberless industrial products (castor and other oilseeds).

The general aim of utilization research on both of the two broad categories of farm commodities is essentially the same--to broaden and extend utilization of the commodities and thereby help to stabilize or increase the demand for them. The scientific procedures of research are broadly the same in both areas, whereas the technologies are in many respects different, especially as between the food materials and all the others. The fundamental justification for carrying on a publicly supported program of utilization research on food products follows a somewhat different line of reasoning than the justification for research to extend the utilization of non-food commodities.

Research toward utilization of non-food products may be based on the avowed public policy of assisting farmers threatened with loss of markets as a result of the swift rise of non-agricultural synthetics, as in the case of wool growers; or on the possibility of developing demand for a presently minor crop to the point where it can be grown profitably on a very large scale in order to remove some of the pressure of surplus from other crops--for example, the development of castor and certain other industrial oilseeds which take land out of cotton and feed grain production.

Research on the processing of farm products for food, on the other hand, is justified primarily by its direct benefit to the entire population through improved nutrition and well-being, reduction of economic losses resulting from spoilage and waste, and increased opportunity to find profitable markets abroad. Indirectly, too, advances in technology through food processing research bring about major and desirable shifts in the commodity supply and demand picture for the country as a whole, as for example in the economical conversion of abundant feed grains into broiler-type chickens, marketable in refrigerated or frozen form throughout the nation and stabilization of dehydrated alfalfa that makes it suitable for export.

Organization of the Division

Research and development along these diverse lines are carried on for the Western Division by a staff headquartered in the Western Regional Research Laboratory, Albany, California. A smaller Department-owned laboratory is operated in Pasadena, California; laboratory space and facilities in Prosser and Puyallup, Washington, are utilized through a cooperative arrangement with Washington State University, Institute of Agricultural Sciences; and laboratory space and facilities in Honolulu, through a cooperative arrangement with the University of Hawaii.

The Albany research staff is organized into six commodity-oriented Laboratories (Cereals, Field Crops, Fruit, Poultry Products, Vegetables, and Wool and Mohair); two functional Laboratories (Pharmacology, and Engineering and Development); and a Pioneering Laboratory concerned with basic studies of plant enzymes. The staff at Pasadena is organized as the Subtropical Fruit Laboratory. The Western Regional Research

Laboratory, at Albany, also houses the Division Director's staff, the staff required for Administrative Management of the Division, and that responsible for Plant Management--that is, operation of the buildings, facilities, and grounds.

Division scientists and engineers not only conduct or supervise research in their own experimental facilities, but also greatly extend the scope and influence of their work by planning and supervising developmental activities carried on by cooperating private firms, processor organizations, or industry groups, and by arranging for research by well-qualified scientists elsewhere under research contracts. In addition, certain grants of research funds are placed with investigators in foreign countries; the cost of these foreign research efforts on behalf of American agricultural interests is borne by Public Law 480 funds.

Examples of Recent Accomplishments of the Western Utilization Research and Development Division

Bulgur Wheat Production Continues to Increase. Making use of information gained in Department research, eight companies have undertaken production of bulgur during the past two years. Altogether 12 companies now produce this highly nutritious food. One of the largest producers uses a new process developed recently by Department scientists. Most bulgur currently produced is purchased for overseas use by the Agency for International Development. Bulgur for this program is made exclusively from Commodity Credit Corporation-owned wheat. Other bulgur made from private wheat stocks is purchased for use by voluntary relief agencies both at home and overseas. Additional bulgur is produced for our regular domestic market, and for Title IV sales abroad for dollars on long term credit. Some five million pounds were sold under this Title in the past year. Purchases during the past two years have increased from an annual rate of 60 million pounds to over 300 million pounds. If the 62 million pound monthly rate at the start of the current fiscal year is sustained as an annual rate, production capacity will have to be again increased from its present 1/2 billion pound level.

Stabilized Alfalfa Meal; A New Export Item. Department research on stabilization of important nutrients of dehydrated alfalfa is playing a vital role in the development of a new export market for this product. Dehydrated alfalfa meal is used primarily as a concentrated source of carotene (provitamin A), vitamin E, and xanthophylls (the pigments which cause the yellow color in egg yolks and in the skin and shanks of broilers). These alfalfa nutrients are rapidly lost by oxidation under ordinary storage conditions. U.S.D.A. research on stabilization of these sensitive nutrients has already led to the development of the antioxidant feed additive, ethoxyquin, which reduces losses to one-third that ordinarily suffered. Without its use, it would be impossible for U.S. producers to

deliver a high potency product in Europe or Japan because of the prolonged "in transit" times required. Since ethoxyquin's acceptance by the Food and Drug Administration in 1958, exports of dehydrated alfalfa have risen from virtually none to 150,000 tons last year. This figure is expected to increase in the future. In the domestic market last year, ethoxyquin was used on over a million tons of dehydrated alfalfa meal and helped the alfalfa dehydration industry maintain healthy growth despite the pressure of stable synthetic competitive products.

Castor Bean Research Leads to New Test for Human Allergies. Allergenic substances present in castor bean meal make it dangerous for humans to handle and limit its use to that of a low-valued fertilizer. Since the castor bean is about half meal and half oil, the low price received for the meal is hindering the growth of the domestic castor bean industry. In research to identify and eliminate or deactivate these allergenic substances, Department scientists have developed an allergy test that uses monkeys as test animals where previously the dangerous and costly use of human volunteers was necessary. Two major risks are involved in allergy testing of humans: (1) the subject may suffer serious anaphylactic reaction to the test allergen, or, (2) he may become sensitized to substances he would not otherwise have encountered. The new test provides a basis for solving problems caused by allergen sensitivity not only to castor but also to other substances. Beyond this immediate objective, the new test has implications in a broad range of scientific research, including physiology, medicine, and phylogenetic investigations.

New Dried Fruit Preservation Method. High-moisture dried fruits are now extensively preserved with sorbates as the result of cooperative research by Department and California Experiment Station scientists, partially supported by industry funds. In recent years there has been an increased consumer demand for high-moisture products because they are more tender and palatable when eaten out of hand, and can be cooked rapidly without prolonged simmering. Because high-moisture dried fruits are susceptible to mold and yeast spoilage, some type of antimycotic treatment is necessary. Sorbates were found to be highly satisfactory for this purpose and conditions for their use were developed. Sorbates do not evaporate even after the package is opened and are effective until the fruit is eaten. Within two years of discovery, the new treatment achieved widespread acceptance, as indicated by the scale of commercial use. Nearly all high-moisture dried prunes and over 50% of the dried figs for market are now treated with sorbates.

Post-Mortem Metabolic Changes Related to Poultry Tenderness. Department scientists have shown that the rate at which glycogen breaks down in muscle to yield lactic acid influences the tenderness of poultry. Undue

stimulation of the muscle at or soon after slaughter (for example, by excessive scalding, beating in pickers, and electric shock) accelerates glycolysis and the onset of rigor. This leads to abnormal toughness. However, if glycolysis is avoided by use of chemical inhibitors or by pre-slaughter injection with adrenalin which exhausts the normal glycogen reserve, birds are tender without the 6- to 12-hour aging period that is usually required. Thus, the occurrence of glycolysis was found to be a cause of toughness. Aging permits poultry to become tender again, the degree of tenderness depending on how fast glycolysis has occurred. This better understanding of post-mortem chemistry of poultry muscle should lead to more efficient methods for producing optimum tenderness in poultry products.

Area 1 WHEAT AND BARLEY-- FOOD AND FEED PRODUCTS AND PROCESSING

Problem. In recent years huge stockpiles of wheat and barley have accumulated, depressing the agricultural economy through restricted grower incomes and expensive government control programs. The most promising solution lies in greatly expanded exports to meet the urgent food needs of large segments of the world's population and to secure an increased share of dollar markets for these grains. Knowledge and skills do not now exist to reach these goals. Ways must be found to adapt U.S. winter wheats and flours to the specific use requirements in Western Europe, a large potential dollar market. New food products from wheat must be created to fit specific needs and preferences of individual countries throughout the world. Simple, inexpensive methods must be devised for use in developing countries to process U.S. wheats into products appropriate for their socio-economic structures. Greatly expanded scientific knowledge of the composition and processing properties of wheat and barley is necessary. More complete knowledge of the chemical and physical properties of both the major and minor constituents of the grains, and of the changes that occur among them during processing, is needed to point the way to the new food and feed products and to new processing technologies. A thorough exploration must also be made of the inherent versatility of these grains as food and feed substances to achieve the utmost of their wide use potentialities.

USDA PROGRAM

The Western Utilization Research and Development Division conducts a broad program of basic and applied research on wheat and barley at Albany, California; under contract at Pullman, Washington; Lafayette, Indiana; Corvallis, Oregon; Kansas City, Missouri; Chicago, Illinois; and Manhattan, Kansas; and under P.L. 480 research grants in England, France, Poland, Italy, and Israel.

Basic studies are concerned with characterizing the soluble proteins (albumins and globulins), gluten proteins, lipoproteins and lipids in wheat and flour, identifying interactions in and between these substances, and characterizing the biologically-active compounds present in bran and germ. Different varieties and classes of wheat are being studied to determine intrinsic differences between the scarce high-quality bread-baking wheats and those surplus wheats which require chemical treatments or aging of flour for bread production. Applied research is being conducted on new and improved food and feed products and processes, with emphasis on the development of products to help fill the food deficit in overseas countries; microbial contaminants of flour and their elimination as a spoilage source for formulated foods; and on the extension of basic research findings to the improvement of manufacture of bread and other baked foods.

The Federal program of research in this area totals 46.1 professional man-years, including three scientists whose salaries are provided by two cooperators under Memoranda of Understanding (Farmers Co-Operative Commission Company - 2, and the Kansas Wheat Commission - 1), and eight contracts providing research at the rate of approximately 5.6 professional man-years per year. Of this number, 22.7 are assigned to investigations on chemical composition and physical properties; 21.7 on new and improved food products and processing technology; and 1.7 on new and improved feed products and processing technology. Research on emergency foods is conducted with funds transferred from the Department of Defense in an amount equivalent to approximately 2.0 professional man-years. In addition, the Division sponsors 13 research grants under P.L. 480 including 10 on basic studies and 3 on applications of research.

REPORT OF PROGRESS FOR USDA AND COOPERATIVE PROGRAMS

A. Chemical Composition and Physical Properties

1. Protein Interactions. The value of wheat for bread-making depends upon the amount and quality of protein it contains. The production of a high-quality bread involves a working of the dough mixture to a proper degree, forming the loaf, and baking the bread. The mechanical development of a bread dough actually is a key to its utility. The work performed on a paste of flour and water transforms it into a smooth strong dough that is both elastic and flowing. To form a good loaf the dough must be elastic enough to retain its gas bubbles, but at the same time, must yield to the pressure of these bubbles to expand. For a product so common as bread, surprisingly little has been understood of the physical and chemical actions involved in the development of doughs for bread manufacture. Accordingly, our research is directed in part towards unraveling these mysteries.

The increase in protein solubility with mixing seems to offer an approach to understanding compositional influences on bread quality. Solubility increases as a result of breakdown of intermolecular cross-links and by molecular fragmentation. When enough of this occurs, the dough loses its strength and volume of baked bread is reduced. Factors that affect the rate and extent of protein solubility during mixing are now under intensive investigation. The mechanisms by which the desirable balance of elasticity and flow properties that are essential to the development of good loaf volume in bread doughs were found to involve the sulfhydryl and disulfide groups of wheat proteins. As doughs are mixed the protein solubility increases; the rate and extent of the increase can be influenced by chemical blocking of the sulfhydryl groups so they can not react. Chemicals that block the sulfhydryl groups were added at equivalent concentrations and a large and rapid increase in protein solubility was observed. When only half enough of the blocking material was added after a preliminary mixing of the dough ingredients, rather than adding it to the ingredients before mixing, the response was more rapid. The sulfhydryl

content had to be decreased to a critical level slightly below one-half the original content to give a large permanent increase in extractable protein.

2. Wheat Flour Dough Rheology. Basic studies toward relating flow characteristics (rheology) to the properties of wheat flour dough, are being conducted under Public Law 480 in the Rheological Laboratory of the Israel Institute of Technology at Haifa. Standardized testing of flour for bread making quality includes the use of equipment such as the Farinograph and the Extensograph. However useful this equipment is, their results have not provided unequivocal predictions of baking quality. Research under this project seeks to identify the basic mechanical parameters which change bread quality, and work out ways of measuring such parameters. A search of the literature was conducted, and exploration made into research conditions and techniques used to study the rheological properties of bread doughs. Equipment was installed, and, in some cases, re-designed to be more appropriate to the study of wheat flour doughs. In preliminary studies, measurement of dough sampled from the three characteristic stages of the Farinograph cycle is yielding information on typical high-quality wheat flour. Experimental conditions are being defined. For example, research must be limited to a very narrow temperature range if uniform results are to be obtained. Furthermore, partially-mixed dough will recover some of its original characteristics when allowed to rest and such rest periods may obscure data on rheological properties for the state of the dough being investigated. The characteristic softness of wheat flour dough has made it difficult to measure pieces of extruded dough in typical rheological measurements. Consequently, equipment is being modified. This investigation should improve our understanding of changes in the physical nature of dough during the mixing process.

3. Analysis and Characterization of Wheat Proteins. The differences between flours seem to depend in large part on proteins and their changes during the development of bread doughs. Consequently, refinement of measurement of types and kinds of protein constituents and changes of proteins during dough development are under intensive study. Pioneering research on immunochemical techniques is being carried out under P.L. 480 at the Pasteur Institute in Paris, France. The techniques of immunochemistry and gel electrophoresis were combined to give an exceptionally good analytical separation of proteins. The separated proteins are identified by the immunological reaction occurring when the antigenic protein combines with a specific antibody present in immune animal sera introduced into the gel. If any separated proteins are enzymes, their activity can be detected in the gel by means of specific color tests. Significant progress has been made in separating and identifying a number of wheat proteins and studies are in progress to relate the amounts and properties of these components to differences in baking properties of different varieties of wheat.

Results of this work are being applied without delay in the Department's research laboratory at Albany. Electrophoretic separations into bands of

protein constituents identified by immunochemical reactions can indicate the complexity of proteins in wheat flour. Polyacrylamide gel electrophoretograms have separated a larger number of components than were detected on agar gels in preliminary immunoelectrophoretic examination of flour extracts. Possibly some compounds separated by the former method have identical immunochemical response or may be polymers representing two or more separated components all having the same response. A procedure combining the resolving capacity of polyacrylamide or starch gels with favorable diffusion characteristics of agar gels has been developed and is being evaluated. Two-dimensional gel electrophoresis has also been undertaken to confirm the identity of component bands in one-dimensional separations. Research is planned in which individual proteins will be separated out and added back to bread doughs to determine their contribution to quality. Preliminary large-scale preparations, up to 35 to 40 grams of protein at a time, have been undertaken using column chromatography.

The development of quantitative measurements for specific proteins is being conducted under contract at Washington State University. A single extraction using aluminum lactate buffers at pH 3 has given the most satisfactory results to date. Such extraction followed by separation with disc electrophoresis appears to be a fruitful approach to quantitative determination of wheat flour proteins.

Another approach to investigating the chemical and physical properties of wheat gluten is being made by careful study of the alterations induced by ultrasonic vibration in research conducted under P.L. 480 at the Institut National de la Recherche Agronomique in Paris. At this stage of the research, it is not clear that reproducible controllable changes in gluten can be effected by the treatment. Further results and evaluations will be conducted, including investigations on gluten extracted from flour by different procedures. When and if controllable changes can be induced, other work will be undertaken to learn of the effects of treated gluten on bread making.

Research on solubility of wheat gluten proteins is being conducted under P.L. 480 at the Centre Nationale de la Recherche Scientifique in Montpellier, France. These studies, intended to provide knowledge of the foaming and surfactive properties of gluten proteins and of the means of increasing and decreasing solubility of these proteins in neutral solution, have been interrupted because of difficulties in obtaining equipment. No progress has been reported yet, and the grant has been extended for two years to enable the research group to complete the work.

In addition, basic research on nitrogenous compounds of wheat germ is being supported by a P.L. 480 grant at Universita di Bologna, in Italy. Data on protein and free amino acids confirmed previous published reports. Significant amounts of polyamines were found in wheat germ. Because polyamines can form complexes with nucleic acids and have physiological significance, they are currently of considerable scientific interest.

4. Chemical Basis for Cohesiveness in Gluten. Basic research on the properties of gluten which contribute to the usefulness of wheat flours in food and industry and to a greater understanding of how new applications may be devised is being conducted by contract with the Midwest Research Institute in Kansas City, Missouri. This project has just recently started. Stocks of crude gluten and purified gluten and glutenin have been prepared. Procedures for chemical modification, such as acetylation, esterification, breaking of disulfide bonds, and blocking of sulfhydryl groups, have been applied to gluten and some modified materials have been prepared. Analytical procedures for determining degree of modification also have been worked out. A procedure for rehydrating the protein specimens with good control of water content, pH, and salt content, and the preparation and mounting of rehydrated gluten strips in an Instron tester has been successfully conducted. Tensile strength, stress, and relaxation determinations will be made on the dehydrated protein strips. A statistical design for this research, covering selection of treatments and modifications of gluten samples, has been submitted and revised on the basis of comments from the subject matter specialist.

5. Enzymes in Wheat and Flour. Studies are under way to isolate and characterize amylase, proteinase, and lipoxidase enzymes known to occur in wheat and wheat flour. Enzymic action is involved in the modifications that take place during the mixing in development of bread doughs. It is also almost certainly involved in the maturation of wheat flours. A systematic study of the enzymes in wheat and the differences in activities of wheat samples may provide some helpful guidance for better means for converting hard red winter wheat into improved bread flour. Four distinct component bands, which have amylolytic activity, were separated by electrophoresis. The standard method for determining proteolytic activity in flours was shortened by several modifications in a new analytical procedure. Sulfhydryl blocking reagents caused a partial inhibition of the proteolytic enzymes and introduction of disulfide and sulfhydryl containing substances into the reaction did not restore the activity.

Studies of flour proteinase indicated that about 25% of the enzyme required a free sulfhydryl group to be reactive, whereas the remainder did not. Lipoxidase activity was found in water extracts from flour, but no activity has yet been found in fractions separated by ion exchange chromatography. Lipoxidase activity was completely lost by boiling the extracts, but sequestering reagents caused no diminution.

Basic studies on the relation of sulfhydryl groups to the amylolytic and proteolytic enzymes in wheat, flour, and malted wheat are being carried out under P.L. 480 at the University of Poznan in Poland. Research there confirmed the importance of maintaining the integrity of sulfhydryl groups in preserving the activity of amylolytic and proteolytic enzymes in wheat. Such enzyme activity is important, and supplemental additions of enzymes to flour are commonly made during milling and formulation of ingredients for

baking. Demonstration of the importance of sulfhydryl groups in preserving enzyme activity dictates establishment of processing conditions such that sulfhydryl groups will be undamaged.

Investigations were initiated on the coenzyme role of riboflavin of wheat endosperm at the College of Agriculture at Poznan, Poland under a P.L. 480 research grant. Preliminary work included development of standardized biometric procedures and separation procedures for wheat constituents important to the study.

In addition, studies of enzyme action in solid natural products particularly in relation to water contents in the range occurring in cereal grains, are being carried out under P.L. 480 at the Institut National de la Recherche Agronomique in Paris. Emphasis is on cereal lipases and studies involving kinetic investigations of enzyme activity. Synthetic substrate mixtures were prepared and dehydrated under conditions of minimum enzyme reaction and kept under well-defined conditions of water vapor pressure. Using isolated lipase from cereal sources, kinetic studies of enzyme activity are being conducted and reaction products will be isolated and characterized.

6. Flour Maturation. Investigations were initiated to develop methods for improving the baking performance of winter wheat flours without the use of chemical agents such as bromate, iodate, or chlorine dioxide and avoiding the expensive holding period for maturation. Preliminary stages of the work are underway. Baking tests were carried out to select suitable flour varying in response to oxidating maturing agents. The lipid components of these flour samples will be isolated and characterized. Comparisons in the composition of spring and winter wheat flour lipids were initiated. A contract has been negotiated to initiate research at Kansas State University at Manhattan, Kansas on qualitative and quantitative differences in the proteins and lipids of hard red spring and hard red winter wheat that account for the characteristically larger response of winter wheat to oxidative maturing agents. Spring wheat responds less to oxidative agents but performs well without treatment. Research in this area is needed to overcome the ignorance of why these wheat flours behave so differently.

7. Lipids and Lipoproteins. An understanding of the nature and function of gluten in wheat flour products requires fundamental information on the chemical and physical properties of their lipoproteins. These form a major class of constituents of gluten and exert an important influence on its properties. Lipid constituents of the lipoproteins of wheat were extracted and segregated into simple, glyco- and phospholipids. Composition of the separated constituents was ascertained by column, paper, and thin-layer chromatography and by other chemical analyses. Comparisons were made between lipid fractions from different types of wheat, and from wheat and wheat fractions that were treated in various ways. Appreciable differences occur in the ratios of glycolipids to phospholipids between flour and gluten and among air-classified flour fractions, depending in part on the protein content of the samples. Despite a general similarity between

the lipids of hard red spring and hard red winter wheat flours, a small but definite quantitative difference was observed. Hard red spring flours contained higher percentages of simple lipids than did the hard red winter flours. The compound lipids of hard red spring flour contained more lecithin and ester groups and less sugars than did the compound lipids of hard red winter flours. The association of lipids with proteins was found almost exclusively within the glutenin fraction of the proteins. Fractionation of gluten by several methods consistently resulted in concentration of lipid in the less soluble glutenin fraction. Lipid appears to prevent some chemical associations of proteins. When gluten lipids were extracted with water-saturated, normal butyl alcohol, the gluten exhibited decreased solubility in dilute acetic acid. A decrease in solubility appeared to reflect an increased association tendency among the glutenin components in the absence of lipid rather than an irreversible denaturation of the protein. Based on this information, more extensive evaluations of differences in flour lipid compositions and lipoprotein characteristics are planned. Selected lipid material will be prepared and compared with natural components by studying effects on dough properties. Although only a small amount of lipid material exists in wheat, it seems to have a very important effect on baking quality.

Work in this area is strengthened by two research grants under P.L. 480. A basic study of the composition of lipids of whole wheat has been undertaken at the Ecole Francaise de Meunerie in Paris, and another grant on studies of phosphorous compounds in flours at the Institut National de la Recherche Agronomique, also in Paris. Research in the first of these projects is concerned with evidence that certain types of fatty materials in wheat are important in determining the properties of flour and thus of a loaf of bread. Lipids undergo changes during preparation of the dough and during yeast fermentation. The results of these changes are being identified.

In the second project, extensive compositional studies have been conducted, comparing 10 United States wheats of different processing characteristics with three French grown wheats representing two of the general types of U.S. wheat. The relationship between total protein and sedimentation value of wheat seems to involve the amount of lipid available. The ratio of total protein to total lipid parallels the sedimentation value. The ratio of lipids left unextracted also seems to parallel the baking characteristics of the flour samples.

8. Bread Flavor. The importance of understanding the chemistry of bread flavor lies in the problem of preserving the aroma of freshly baked bread. By understanding what flavor is, and how to measure it, we start toward flavor enhancement and flavor stability. The exceedingly painstaking task of determining minute components of bread that create its delightful flavor involves development of elegant and sophisticated equipment and procedures to measure nerve-stimulating materials in fractions of a part per million.

The ingenuity demanded is matched or exceeded by that required for the second task of correlating compositional data with flavor and aroma responses. Organoleptic evaluations and correlation of subjective response to individual components will be emphasized. Eighty to 100 components of bread have been isolated and identified in volatile forms. These volatiles are extracted from cooked breads, or captured from the oven vapors while bread is being cooked. One fraction, which has a characteristic fresh bread flavor, was isolated from the oven volatiles. When exposed to air, it developed a typical stale aroma. A part of the investigation is directed to the volatile components produced in pre-ferments, such as are used in the continuous mix manufacture of bread. Nine different keto acids were found to occur in significant quantities in pre-ferments used for bread production. Tentative identity of these nine compounds has been made and analytical methods are being developed to measure them quantitatively in pre-ferments of various composition and history. With the knowledge being gained,each keto acid can be added in various amounts to pre-ferments under controlled conditions and its effect on flavor and aroma determined.

Related contract studies at Massachusetts Institute of Technology have been completed. In this contract research the effects were measured of modifying the composition of pre-ferment bread doughs on the volatile flavor and aroma components of breads baked therefrom. Taste panel comparisons were made of pre-ferment breads that had been prepared with additions of individual amino acids. Several of the amino acids produced significant changes in flavor and aroma. However, proline was the only additive which produced a bread preferred over controls. Quantitative comparisons were conducted on volatile materials collected from freshly baked samples of control bread and bread made with proline added to the pre-ferment. The addition of proline increased the total oxidizable material of the volatile concentrates. The increase in volatile concentrates was, within limits of measurement, accounted for by increase in ethyl alcohol in the volatile material collected. Concentrates prepared from volatile distillates of the control bread and proline containing bread were not found significantly different in chemical composition despite the organoleptic differences that had been found. Forty-one compounds were isolated from bread volatiles and 24 of these were identified, including 9 which had not been found in bread before.

There have been many significant advances in recent years in the chemistry of bread flavor and the tools of research have advanced so that progress should accelerate. However, there is still a long way to go before the chemistry of bread flavor is so well understood that we can adjust the ingredients and processes of bread-making to obtain products of superior flavor that are resistant to staling.

B. New and Improved Food Products and Processing Technology

1. Bulgur and Related Wheat Food Products. Bulgur, or parboiled wheat, has been continuously used as a diet staple since post-neolithic times in the Near Eastern cradle of civilization. Although not a substantial commercial item in the United States until a few years ago, bulgur is among the most important agricultural exports of the government donation program. In 1963 about 300 million pounds were shipped under Public Law 480. In a pilot program of the domestic school lunch distribution 800,000 pounds were used. Obviously as bulgur becomes more widely known, sales of wheat in this form will increase. A commercial venture in bulgur manufacture initiated during the past year, utilizes a process developed by the Department of Agriculture. This plant can produce 10 million pounds of bulgur per month and brings additional capacity to meet the rapidly growing demand for this product. The Department-developed process involves continuous conditioning and cooking at atmospheric pressures. The process uses either red winter wheat or white club. Bulgur obtained from both wheats was highly satisfactory judged by milling yield, organoleptic quality, and nutrient content.

Several canned bulgur products were produced in 150 to 200 can lots to evaluate formulations and processing, and to provide supplies for demonstration purposes. Cracked and whole kernel bulgur generally performed more satisfactorily than peeled raw wheat in canned products, but slight changes in formulation were necessary in replacing the raw wheat with the bulgur. Preliminary trials with rotating or tumbling pressure retorts indicate that it will be feasible to fill cans with the dry materials and blend the ingredients by agitation during processing. Products made this way were more uniform than mixed products cooked in a still pressure retort.

Quick-cooking bulgur was developed that provides for a wide variety of instant consumer products from wheat. Hot air puffing of whole kernel bulgur transforms it to crisp, expanded structures, which quickly absorb hot liquids so formulations can be made that are ready-to-serve in a matter of minutes. Instant soup, salad, desserts, and other mixtures were made to demonstrate the puffed bulgur.

A wide variety of attractive and nutritional wheat foods has been developed and recipes prepared and widely distributed. Demonstrations of many of these foods have been made to interested associations of wheat growers and professional and social organizations. Assistance has been given in the development of recipes suitable for the use of bulgur in the school lunch program.

2. Food from Wheat Fractions. New canned and dehydrated high-protein food products that utilize wheat are being investigated. Studies include work on technology to modify structural and textural properties of products made

from flour with and without protein fortification. The unique properties of starch components of wheat may be developed to make desirable textural changes. Protein-rich fractions obtained by air classification of flour and from mill feed fractions, are being looked to in the formulation and development of more nutritious products. Canned products using gluten as a binder, which are flavored and treated to suggest poultry, fish, and beef, have been produced in preliminary work.

Acid hydrolytic deamidation of gluten to improve wheat protein dispersibility in water and buffered solutions was demonstrated in contract work at Purdue University, Lafayette, Indiana. A product completely dispersible in a dilute phosphate buffer was produced by treatment of gluten with dilute hydrochloric acid. Twenty percent of the amide nitrogen was split from the protein and some peptide bonds were also hydrolyzed. Phosphoric acid hydrolysis may prove to be more effective and studies are being extended to air-classified high-protein flour fractions as well as gluten. Preliminary work shows promise in the development of powdered formulations of wheat that contain minerals and vitamins and would be usable as a food supplement in milk-short areas of the world.

3. Emergency Food Supply. Investigations of foods suitable for fallout shelters are being conducted with funds transferred to Agriculture by the Department of Defense. Two lines of research are involved: (1) on the bulgur wheat wafer previously conceived by the Western Utilization Research and Development Division for stockpiling in fallout shelters, and (2) on development of adjuncts for use with the bulgur wheat wafer. Close cooperation was given by Department scientists to the Office of Civil Defense of the Department of Defense and the Van Brode Milling Company of Massachusetts in connection with a half million pound trial procurement of the bulgur type shelter wafer.

Stability of bulgur wafers is being investigated in a five-year storage test recently initiated by a research and service contract with Oregon State University in Corvallis. Variables in the study include red and white wheat as the basic material, malt and corn syrup, nitrogen vs. air pack, storage at 40, 70, and 100° F.

Study of alternate methods for preparing expanded wheat is in progress. Heat-puffing steamed wheat without the normal bulgur preparation did not provide enough expansion of the individual kernels to produce a satisfactory wheat wafer. A cereal puffing gun has been installed for further studies. Hot fat puffing of debranned raw wheat was explored. Debranned white wheat of different moisture levels was treated in hot fat over a wide range of temperatures. Oil-puffed wheat had a pleasant toasted flavor but the puff indices indicate that further improvement would be necessary for good texture in the final wafer.

Efforts have been directed towards quicker evaluation of storage stability. Irradiation of ground puffed bulgur with ultraviolet light has been found

extremely effective in accelerating the changes measured by gas-liquid chromatographic patterns. If the hexanal produced is used as an indication of change, samples of ground puffed bulgur exposed to ultraviolet light in air showed increases of about 25-fold in eight days, 50-fold in nine days, and 75-fold in ten days. Exposure in oxygen in the dark, showed only a 3- to 4-fold increase in ten days.

Wafers have been made with substances reported to protect against radiation or radioactive fallout. Calcium carbonate, tribasic calcium phosphate, ascorbic acid, L-cysteine hydrochloride, potassium iodide, yeast, folic acid, methionine, and sodium calcium citrate have been added and the levels determined at which there is no adverse effect on flavor, texture, or general appearance. Should such additions be indicated, the quality of the wafer would not be affected. Possible influence of these additives on wafer stability is yet to be discovered.

Foods that would be useful with emergency supplies of bulgur wafer were selected for extended storage tests. Such tests were initiated by a second contract with the Oregon State University at Corvallis.

Considerable success has been achieved with cold water gels as food adjuncts for bulgur wafers. Further improvement in the clarity, flavor, and ease of mixing is being sought. Such products could be stored dry for stability, mixed with cold water, and used as flavored spreads for the wafer to provide variety. Investigations have been undertaken also on a highly stable fat spread resembling margarine that could be used with the wafer. Preliminary work is well under way on a different package for dry mixes which may be reconstituted with cold water (e.g., spreads, cold water gels, and icings). The dry mix would be packaged in an oversized plastic bag within a tin can. The bag can be removed from the can, cold water added, and mixing effected by kneading the bag. The adjunct could be extruded from the bag through a hole produced by cutting off a specially designed tip. Adjuncts packaged this way would require no equipment for preparation and would go directly onto the wafers.

4. Baking Quality. Baking quality of wheat flour, as related to compositional factors, is under continuing investigation. Air-classified fractions are now available over a wide range of protein content from five hard red winter wheat varieties. Bread baking and physical mixing tests of dough are in progress to study relationships of protein quantity and quality to baking quality. Unclassified flours are being sought which are similar in protein content to the various air-classified fractions. These natural flours will also be evaluated for baking quality to see if results obtained are the same as those in the separated fractions.

The effects upon baking of variation in wheat flour lipids are being investigated under P.L. 480 at the British Baking Industries Research Association in Chorleywood, England. Representative samples of U.S. grown wheat

are being analyzed for fat, evaluated for baking quality, and studied with formula modifications to determine the role of fat in bread loaves. Because varietal and cultural factors influence quality of samples, several years of investigation must be evaluated before conclusions can be drawn. However, even at this stage in the research, evidence is accumulating that certain fats are indeed important to the behavior of dough, the loaf volume, and crumb firmness of bread.

Continuous-mix bread manufacture has brought up a new problem in making bread doughs. Some flours tolerate nonfat dried milk to a greater extent than others. Research was initiated to study what components in flour limit the use of nonfat dried milk in continuous-mix bread manufacture. Tolerant and non-tolerant flours were selected for this study, and research was initiated to determine the relationships between milk solids and individual components of these flours. By cooperation with the Eastern Utilization Research Division, milk solids that have been treated in various ways are available for dough formulas to study their effects on baking quality. Specific changes occurring in the flour during mixing and baking will be examined. The objective is to develop methods for selection of flours with high tolerance, to determine milk treatments which will overcome the shortcoming, and to develop variations in baking procedures that will permit the usual amounts of milk solids in bread formulas.

5. Nutritive Value of Processed Wheat. The application of heat during the manufacture of bulgur and other wheat products, raises the question of nutrient retention. However, vitamin losses were found minimal for a range of conditions representing commercial operations. Preliminary chick growth experiments indicate that biological protein values are as good as or better than those of flour made from the same wheat. Leaching of protein from the bran into the endosperm may make amino acids more available. The nutrient quality of bulgur is approximately that of whole wheat flour, with thiamine losses less than 15%. Thiamine losses during the canning of wheat and the puffing of bulgur were significant, and the puffing operation somewhat impaired biological protein value. The processed products are nevertheless fair to good sources of several B-vitamins, including thiamine. The losses do not exceed the losses that occur in toasting and cooking of many common wheat products.

Studies aimed at rapid chemical methods for assay of nutritive value of proteins during processing of wheat products are being conducted under P.L. 480 at Cambridge University in England. Compositional studies relate the effects of heat to amount of lysine, methionine, and leucine in wheat products. This information is being related to the biological value of proteins containing these amino acids. These studies correlate data from rapid chemical analyses with those from the time-consuming animal assays. If these chemical tests correlate adequately with nutritive value, it will be possible to monitor industrial processes used in the production of wheat food products.

6. Elimination of Microbial Contamination of Wheat Flour. The recent growth of convenience foods, such as frozen and canned products and premixes with flour, has brought attention to micro-organisms that frequently contaminate flour but that are adequately controlled when flour is used in commercial bread or other bakery products. Sanitation requirements are becoming more stringent in frozen foods, baby foods, and other products and have caused food manufacturers to limit wheat flour in many products. Refined starch, purified vegetable gums, and other thickeners are used in place of wheat flours for better sanitary control. Contract research at the American Institute of Baking in Chicago has been negotiated to determine the nature and extent of contamination that would be detrimental to flavor, quality, and safety of foods containing flour and to develop methods to eliminate contamination from flour during processing. Future research will include studies on the viability of organisms in flour for various uses and the effects of processes to reduce microbial contaminations to a negligible level. Research on this contract will begin in the very near future.

C. New and Improved Feeds and Feed Processing Technology

1. Improved Feeds from Wheat and Wheat Fractions. Research on feed utilization of wheat has been limited during the period of this report by unavailability of personnel. Chick feeding tests were conducted to determine the effects of bulgur processing on the nutritive value of wheat. Studies were made of both hard red wheat and Northwest club wheat. The bulgur process does not reduce lysine availability. Research in this area will develop toward new feeds based on the high-protein wheat milling byproducts. These materials contain higher levels of protein than the flour or the whole of the starting grain. Furthermore, the middlings, shorts, and other parts of the mill run have protein that is of greater biological value than that in the flour. Representative samples of mill run will be separated and fractions characterized for amount and biological quality of protein. Wet and dry separation and combinations thereof will be used to prepare protein-rich fractions, which because of concentration and biological value, would be useful to the feed mixing industry.

Contract research on estrogens in wheat bran and germ conducted at Iowa State University, Ames, has been concluded. Oils from wheat bran and germ were fractionated. Various fractions were assayed biometrically for estrogen-like activity. Such activity had been previously reported in the hydrocarbon fraction and was confirmed. Another fraction appeared, in preliminary bioassay procedures, to be of higher activity than the mixed hydrocarbons and obtainable in good yield. Research concentrated on this apparently homogeneous phenolic substance. On detailed study, it was found to be a homologous mixture of five compounds varying slightly in the number of carbon atoms on the side chain of an alkyl-dihydroxy benzene. Subsequent bioassay of the purified components did not bear out the original finding of estrogenic activity. It must be assumed that impurities not yet revealed but removed in the purification were responsible.

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Area 2 RICE--PROCESSING AND PRODUCTS--WESTERN LABORATORY

Problem. The productive capacity of U.S. rice growers has increased faster than domestic and export consumption over the past decade, limiting the income potential of this major world food grain. Detailed knowledge of chemical composition and physical properties, as related to processing, is needed to guide milling, processing, and product development of U.S. rices so that they can better meet the quality requirements of expanded markets. New and diverse food products from rice that are easy to prepare, have flavor and texture appeal, and are economical to manufacture, are needed to increase the total consumption of rice both here and abroad.

USDA PROGRAM

In the Western Utilization Research and Development Division, basic and applied research on rice is conducted at the Division headquarters, the Western Regional Research Laboratory in Albany, California. Basic studies involve chemical, physical, and biochemical investigations of rice proteins and of processing. The protein work is concerned specifically with isolation and characterization of the globulin and glutelin proteins of the endosperm. Effects of processing on quality of cooked rice are being studied. Preparation of high-protein rice fractions by means of fine-grinding and air-classification is under exploratory investigation.

The Federal program of research in this area totals 3.8 professional man-years. Of this number 2.2 are assigned to chemical composition and physical properties; 1.6 to new and improved food products and processing technology.

REPORT OF PROGRESS FOR USDA AND COOPERATIVE PROGRAMS

A. Chemical Composition and Physical Properties

1. Rice Proteins. Investigation of processing, cooking, and nutritional qualities of rice, leads repeatedly to questions concerning rice proteins. Rice, as now available, is a food that requires protein supplementation to sustain healthy human life, although what protein is available in rice has a very high biological value. The proteins of rice also appear to affect processing and cooking quality. As a consequence, fundamental research on the protein constituents of rice is being conducted. Rice protein has been extracted and separated by solubility differences into globulin, prolamine, and glutelin fractions, which are now being characterized. Electrophoretic patterns of rice globulin have indicated 11 or more components. Rice globulin has been found to have an unusually high sulfur content, reminiscent of β-globulin of barley. No such protein has been found in other

cereals. Further characterization is underway, including studies of the amino acid content of individual proteins.

B. New and Improved Food Products and Processing Technology

1. Cooking Characteristics of Rice. Evaluation of the cooking and processing characteristics of rice is important in selecting raw material and developing new processes and products. An objective method to determine degree of gelatinization, and from that the quality of cooked rice, was developed using the Amylograph. Further studies on rice will include use of the new procedure to evaluate means of processing rice to improve its utility for various markets.

2. High-protein Rice Products. Rice could be more useful, particularly for export to underdeveloped nations, if its normal protein content could be enriched. Substantial increases in rice protein content have been obtained by using high-level applications of nitrogen fertilizers at the California State Experiment Station. Three varieties of rice grown at the Station, which contained 9 to 10% protein, were used in processing studies to develop food products of even higher protein content. Turbo-milled and air-classified rice flours were obtained with 16 to 17% protein in 8% of the flour. The remaining material yielded another 5% of flour containing 11 to 14% protein. In view of the high biological value of rice protein, a rice flour containing in the neighborhood of 14% protein, would be a very nutritional food product and of great value in the protein deficient areas of the world, if this process could be made economical. Profitable outlets for the separated low protein fractions must be found.

PUBLICATIONS AND PATENT REPORTING RESULTS OF USDA AND COOPERATIVE RESEARCH

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Area 3 FORAGES AND FEED-- PROCESSING AND PRODUCTS

Problem. The demand for livestock in the United States will increase 45% by 1975. Fresh forage crops are the richest natural source of many nutrients for farm animals. The bulk of forages, however, is preserved so inefficiently by haymaking and ensiling that 10 to 50% of the dry weight, and much larger fractions of the most valuable nutrients, are lost before the animals eat them. Dehydration is now the only practical means of preserving the high nutritional value. Poultry and swine producers are aware of the value of dehydrated forage, but restrict their consumption because of high fiber and growth-inhibitor content. The livestock breeder needs forage products tailored to specific animals, and the forage producer must adapt to his needs to sell.

Basic and applied utilization research are necessary to produce: (1) nutritional juice and low-fiber feed for non-ruminant animals; (2) fiber products which have been cheaply treated to make them easily digestible for ruminants; (3) growth stimulating supplements for ruminants based on the biologically active fiber digestion factor, and growth-promoting factor in forage. Further, new products should be adaptable to mechanical feeding. Improved handling will encourage farmers to put high value land now producing surplus crops into forages.

USDA PROGRAM

Current research in the Western Utilization Research and Development Division includes both basic and applied studies on forages, principally alfalfa and other legumes. The research is conducted at the Division headquarters at Albany, California, under contract at Berkeley, California, and under the P.L. 480 grant program in Edinburgh, Scotland. Basic compositional studies deal with the potent estrogen coumestrol (discovered by Department scientists), and other phenolic compounds present in forage legumes. The value of coumestrol-rich alfalfa as a growth stimulant for ruminants is being studied with financial support of the American Dehydrators Association. Also under study are other biologically active forage constituents, such as the chick growth-promoting factor in forage juices and alfalfa saponins which depress chick growth, and the mechanism of action of forage antioxidants. Processing of forages by "wet" (juicing) and "dry" (turbomilling and air classification) methods is being investigated.

The Federal program of research in this area totals 14.2 professional man-years, including 2.0 scientists whose salaries are provided by the Department of Agriculture and Inspection, State of Nebraska, and contract research equivalent to 0.5 professional man-years per year. Of this number 6.5 are assigned to chemical composition and physical properties; and 7.7

to new and improved feeds and processing technology. In addition the Division sponsors, under P.L. 480, a research project on forage composition.

REPORT OF PROGRESS FOR USDA AND COOPERATIVE PROGRAMS

A. Chemical Composition and Physical Properties

1. Phenolic Constituents of Forages. Alfalfa and ladino clover are being extracted to isolate groups of compounds and individual compounds that exhibit biological activity. (See paragraph 2.) Chemical identifications are made and sufficient quantities of active compounds are obtained to be used in feeding studies. Under a service contract, 45 tons of alfalfa were extracted with acetone. From the extract, 1000 grams of purified coumestrol were obtained. The residue was further studied and separated by countercurrent distribution. Twelve components exhibiting intense fluorescence under ultraviolet light were isolated. These substances will be further characterized in forthcoming studies. Preliminary findings indicate that they are phenolic and some are structurally similar to coumestrol. Biological activity will be measured to discover whether or not some of these components may contribute to the occasional unexplained grwoth in alfalfa-fed animals.

2. Pharmacological Research on Forages. Biological activity of alfalfa and clover is determined by assay with laboratory animals. Pharmacological tests are made on concentrates, extracts, and fractions. Much attention has been devoted to coumestrol from alfalfa, which exhibits estrogenic activity in mice. Recently, several samples of alfalfa have contained coumestrol up to 200 to 500 p.p.m. with which it is possible to measure the activity of the alfalfa meal rather than an extract. Unexpectedly, when these alfalfa meals were fed to mice, the calculated estrogenic response based on chemical assays was not realized. However, if the acetone extract of the meal, which also contains coumestrol, is fed to the mice, there is a three-fold increase in uterine weight indicating estrogenic activity. Possibly an estrogen inhibitor may be a natural component of the alfalfa meal. In Australian research the isoflavone formononetin was found to exhibit considerable estrogenic potency in sheep. This plant estrogen, when assayed in mice, indicated a relatively low biological activity. A re-evaluation of the mouse assay as a criterion of potency in ruminants must be initiated.

3. Interaction of Forage Antioxidants. A new contract was made with the University of California, Berkeley, to follow leads uncovered under a previous contract. Lipids from alfalfa will be studied, and the chemical effects of processing and antioxidants on lipid oxidation will be elucidated. The investigation will be directed toward separating and characterizing lipid components of alfalfa, identifying changes that occur in them as a result of dehydration and storage, and determining the effects of various antioxidants and the chemical mechanisms by which their effectiveness is accomplished.

4. Structure of Alfalfa Polysaccharides. A structural analysis of alfalfa polysaccharides is being conducted under Public Law 480 funds at the Edinburgh University, Scotland. Research has been initiated along two lines. An examination of the polysaccharide components of alfalfa has been started with extraction procedures designed to obtain individual polysaccharide fractions for further identification and study. Secondly, a survey of the enzyme carbohydrase activity of extracts of dormant and germinating alfalfa seeds, stems, and leaves, has been initiated. Preliminary experiments gave evidence of several enzymes that are active on polysaccharide substrates. Large-scale preparations of these enzymes will be made.

5. Organic Acids. A study on the organic acids in alfalfa was initiated in cooperation with the Kellogg Foundation, whose grant program enabled an English biochemist, J. M. A. Tilley, to conduct research at Albany, California. Special chromatographic apparatus was obtained and details of methodology have been worked out appropriate to alfalfa. A Department scientist will continue independent study of the organic acids in forages based on the preliminary findings of Dr. Tilley.

B. New and Improved Feeds and Processing Technologies

1. Coumestrol-Enriched Feeds. Large cooperative animal feeding trials have continued into the third year using high- and low-coumestrol dehydrated alfalfa, screened from production lots at commercial dehydrating plants. Alfalfa meal for these studies has been obtained with financial help from the American Dehydrators Association. After three seasons, sheep feeding experiments at Oregon State University using alfalfa that is rich in coumestrol have indicated a 20% increase in gain and the production of meat. Meat quality has been judged superior to that from controls. Similar tests with beef steers did not show positive results at the coumestrol levels involved. Samples of coumestrol extracted by acetone from alfalfa have been sent to the Animal Husbandry Research Division for cooperative studies on protein balance in cattle, and to the Oregon and Indiana Experiment Stations for cooperative studies on lamb and steer growth.

2. Improved Alfalfa Meal. Alfalfa meal is being divided into low- and high-fiber products to provide specific components for mixed feeds. This research is supported in part by the State of Nebraska, Department of Agriculture and Inspection, which provides the salaries of two professional employees. Air-classification of fine-ground alfalfa was not effective. A more promising approach involves separation of leaf from grit and stemmy portions and then fine grinding and air-classification of the leaf fraction. Alfalfa containing 26% protein and 17% fiber has been separated into two components, one of which contains 29% protein and the other 30% fiber. Research on the composition of the alfalfa plant indicates 30% to 40% protein in clean leaf fractions and 40% to 50% fiber in pure stem fractions. Therefore, some further enrichment appears possible if more refined separation procedures can be developed.

Stability of dehydrated alfalfa has been improved by the antioxidant ethoxyquin, as reported previously. A substantial export market for ethoxyquin-treated dehydrated alfalfa has been developed. The long hot shipment overseas of alfalfa meal requires a more stable product than untreated alfalfa meal. Without an antioxidant, alfalfa meal imported into Japan and western Europe would have little of its specific nutrient value. Recently the Food and Drug Administration has set a tolerance of 5 p.p.m. for residues of feed additive ethoxyquin in or on the uncooked fat of meat from animals, except poultry, and a tolerance of a half a part per million for residues of the additive in or on uncooked muscle meat.

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Area 4 WOOL AND MOHAIR-- PROCESSING AND PRODUCTS

Problem. Synthetic fibers are making increasing inroads into many of the traditional markets for wool and mohair because synthetics are shrink resistant, quick to dry, wrinkle resistant, and able to hold pleats and creases. Despite the superiority of wool and mohair in tailorability, comfort in wear, appearance, and hand, they lack some of the requirements for ease-of-care performance. Moreover, in present processing practices and in many of their uses, wool and mohair are subjected to conditions which result in damage, distortion, or weakening of the fibers, and in undesired changes in performance and appearance of fabric.

Needed are practical modifications that give durably wrinkle-resistant lightweight wool fabrics; fabrics that are more resistant to soil, acids, alkalies, wear, pilling, and abrasion; fabrics that have greater resistance to felting and relaxation shrinkage; and wools durably resistant to yellowing, to insects, and micro-organisms. Needed also are new types of fabrics, woven and non-woven, for industrial and other uses, made from natural wools, blends of wool with modified wools, and with other fibers. Development of new and improved wool and mohair products and processing methods will require fundamental information on the chemical, physical, and structural nature of these fibers. If a stable sheep and wool industry is to be sustained, mills must be supplied with processing information on how to produce new and better wool products more efficiently. Inroads have been made in wool markets because the uniformity of price and quality of synthetics and the detailed information which producers of synthetics supply for processing these fibers on textile machinery for wool makes them easy and profitable to use.

USDA PROGRAM

In the Western Utilization Research and Development Division, a broad program of basic and applied research on wool and mohair is conducted at the Division headquarters at Albany, California; by contract in Lowell, Massachusetts, Durham, North Carolina, and Washington, D. C.; and by grant funds under P.L. 480 in England, France, and Finland. Fundamental research is conducted on wool and mohair to relate chemical composition and structure, molecular structure, physical structure, physical properties, and surface properties of both normal and chemically modified fibers to the performance characteristics of the fibers in yarns, knitted and woven fabrics, and non-woven forms such as felts. Fundamental research is conducted on the chemical modification of wool and mohair to impart resistance to degradation by heat, light, and chemical environments encountered in use, and to improve use properties such as washability, crease retention, wrinkle recovery, and resistance to staining, abrasion, and insect attack. Applied research is conducted to develop practical processes for the chemical or physical

modification of wool and mohair fibers, yarns, fabrics, and felts; to develop processing procedures for the modified fibers; and to develop new and improved products from the modified fibers; all to increase the utilization of wool and mohair. In addition, Department scientists are making every possible effort to bring research results to the industry through technical publications, public service patents, popular articles, TV and radio broadcasts, participation in growers' and processors' meetings, exhibits, mill visits and development trials, and conferences with visiting mill men.

The Federal program of research in this area totals 41.4 professional man-years, including contract research equivalent to approximately 1.7 professional man-years per year. Of this number, 25.9 are assigned to chemical composition and physical properties; 15.5 to new and improved textile products and processing technology. In addition, the Division sponsors research grants under P.L. 480 including two on basic studies and two on application of research findings.

REPORT OF PROGRESS FOR USDA AND COOPERATIVE PROGRAMS

A. Chemical Composition and Physical Properties

1. Chemical and Molecular Properties. Basic research is continuing for the purpose of better understanding wools to provide an intelligent basis for modifying and processing them into new and superior products. The rupture of wool fiber components has been accomplished with minimum chemical alteration by ball mill disintegration of wool fibers at low temperature. Extensive chemical and physical measurements of wool protein were made and evaluated with the aid of computers to determine the molecular properties of proteins and polypeptides from natural and modified wools. The fiber surfaces of wool and modified wool were studied by the electroosmosis technique, which showed that the WURLANized wool fiber surface has an increased isoelectric pH. Isoelectric changes are reflections of alteration in fiber-to-fiber interactions responsible for felting and shrinking. Certain wool degradation products, resulting from ultraviolet irradiation, were tentatively identified and accounted for by simple molecular fragmentation, or, in some cases, by oxidation. Basic research on wool is continuing.

Nuclear magnetic resonance was used to study water bound in wool fibers. Data are being obtained to further the understanding of the behavior of specific hydrogen atoms of glycine, and will be used as a model to study similar hydrogens present in wool. Hydrogen bonding is responsible not only for binding of moisture by wool, but also for maintaining helically coiled protein structures and for tying these structures together.

Other reagents for modifying wool were investigated. These include hexamethylene diisocyanate, octadecyl isocyanate, and epichlorhydrin. Such reagents tend to improve wool's resistance to acids, alkalis, and oxidizing agents and to impart varying degrees of resistance to shrinkage. Diisocyanates tend to lower abrasion resistance, apparently because of cross-linking, whereas some monoisocyanates do not impair abrasion resistance. Reagents frequently, but not always, reduce dry and wet strength. Grease wool that contains over 20% water yellows significantly within two weeks at 35° C. The yellowing, which increases with time and moisture content, is effectively inhibited by formaldehyde. The formaldehyde reacts with the wool to increase its resistance to acids and alkalis. Work is now in progress to determine the practicality of formaldehyde treatment.

An investigation of the distribution of sulfur in wool was concluded. The research was conducted at the Wool Industries Research Association in Leeds, England, under a Public Law 480 grant. It led to the discovery of bis(2-amino-2-carboxyethyl) trisulfide in wool hydrolysates and cleared up a puzzling discrepancy in the analysis of sulfur in wool. The sulfur compounds of wool serve as components of the polypeptide molecules and also as cross links connecting the molecules and influence shrinkage, crease retention, wrinkling, etc. Known sulfur-containing compounds could account for only 80% of the total content of sulfur in wool. New sulfur compounds were identified which account for much of the missing 20%.

A study of the sequence of amino acid building blocks in proteins of wools is being conducted at the Universite de Lille, Lille, France under a P.L. 480 grant. A low-sulfur protein has been isolated in more nearly pure form (about 50%, judging from analytical results) than hitherto. It is part of the main fibrillar component of the wool fiber. As such, it participates in determining useful properties of wool such as tensile strength and extensibility. The methods applied are being refined and simplified, and research is proceeding to find how the building blocks of this protein fit together.

2. Physical and Mechanical Properties. Basic physical studies are conducted to establish the mechanical properties of natural and modified wool and mohair fibers to assist in the development of new and improved products and processes. The elastic recovery and resilience properties of WURLAN treated fibers and natural wool fibers were compared. The small amount of polymer deposited by the WURLAN treatment does not change these important properties over an extension range of 2% to 30%. Refinements were made in equipment to measure the fineness of individual wool fibers. A constant stress, fixed-frequency vibroscope has been put into routine use for fineness measurement. With this instrument crimped fibers are uniformly straightened and higher precision in fineness measurements is obtained. This represents an improvement over the standard microscopic measurement which is biased toward the major axis when fibers have

elliptical cross-section. It was demonstrated that the fiber fineness and length distribution characteristics of blended top can be predicted from the characteristics of the components of the blend. In routine fineness determinations, results may be accurately estimated by graphical procedures which eliminate all calculations, save time, and reduce errors.

In studies of the torsional properties of wool fibers, removal by heating or chemical modification of the disulfide cross-links, which are responsible for many of wool's unique properties, led to sharp reduction in modulus (stiffness) and increase in solubility of the fiber in formic acid. Such studies of the contribution of cross-links between wool fiber molecules as they relate to mechanical properties of wool under many solvent-temperature conditions provide information that will suggest structural and chemical modifications to improve specific properties of wool.

3. Effects of Radiation and Other Physical Forces on Wool. Progressive changes were demonstrated in the amino acid composition of wool exposed to ultraviolet light for varying periods of time. The effects of ultraviolet light, X-ray, and mechanical damage on wool were investigated by following color changes and electron paramagnetic resonance spectra of induced free radicals. Wool contains a naturally-occurring, stable, free radical in low concentration. In contrast to this natural free radical, the radicals produced by ultraviolet or X-ray are unstable to heat, water, and oxygen. Ultraviolet light can turn wool green, yellow, or white, the color depending upon the wavelength of incident light and conditions of irradiation. By comparing electron paramagnetic resonance spectra of irradiated wool and irradiated individual amino acids, it was shown that the spectrum obtained with green wool could be accounted for by tyrosine and cystine radicals. On exposure to water vapor or air, the green color changed to yellow in a short time, and the free radical spectrum eventually returned to the stable natural radical. Amino acid analyses of wools which have been extensively irradiated with ultraviolet light showed that glycine and alanine increased slightly while all other amino acids decreased, cystine disappearing completely. Several new amino acids appeared as the result of either methylation of demethylation of a naturally occuring amino acid. The end result of irradiation of wool is to turn it yellow. Intermediate colors have been observed only when conditions of temperature, wavelength, intensity of irradiation and gaseous environment were specifically controlled. Research will be continued on the properties of natural fibers to provide basic information on the ultraviolet degradation of wool that will guide chemical modification to improve its performance.

B. New and Improved Products and Processing Technology

1. Shrink Resistance. Development of an interfacial polymerization treatment for wool textiles to impart shrink resistance was reported last year. This new process, covered by public service patents, has been given the name WURLAN. This process has been developed into full-scale mill application and its importance is best expressed by a recent press release

from a trade association during the past year: "A revolutionary new process for shrink-proofing wool fabrics has been developed by the United States Department of Agriculture, enabling wool clothing of all types to be machine washed without appreciable shrinkage." The American Sheep Producers' Council, representing all segments of the American wool textile industry, is assisting in promotion of the new development, in cooperation with the U.S.D.A., as a part of a continuing program to provide information on the process to consumers. One of the largest wool textile producers in the country is now commercially producing WURLANized wool fabrics. Their production rate will increase markedly when promotion begins in the near future.

Initial developments of the WURLAN treatment were applicable to woven wool textiles. A procedure was subsequently developed to apply WURLAN to tubular knit goods but was not satisfactory unless the tube was opened. But because knit goods are subject to distortion in handling, it now appears best to apply the WURLAN treatment to the wool before it is spun into yarn. This development has now been accomplished on laboratory and pilot plant equipment and is being extended toward commercial-scale equipment. The extension of the WURLAN process to wool top and thus to knit goods will help regain markets that have been dominated by synthetic fabrics in recent years.

Treatment of wool by interfacial polymerization has concentrated on utilizing hexamethylene diamine and sebacoyl chloride to form polyamide 6-10. Research is continuing on alternative monomers for this application. Exploratory studies indicated that bischloroformates, which react with diamines to form polyurethanes on wool, may be superior to diacid chlorides in the WURLAN process in stability and in cost. Potentially cheaper than diacid chlorides, C_4 and C_6 bischloroformates are of primary interest. The C_4-diol is commercially available but the C_6-diol will not be commercially available before 1964. Among other polymers being investigated, modified polyethylenes are of particular interest because they provide good shrink-resistance along with excellent fabric hand. They are, however, difficult to apply. Exploratory research was started on new polymers. The oxidative coupling technique provided a rapid polymerization with commercially available phenolic monomers to yield polymers with properties not realized with vinyl polymers or condensation polymers. Preliminary investigations also were initiated on the grafting of collagen onto wool. Because collagen is commercially available and inexpensive, it could be an alternative and cheap finishing agent if grafting to the wool could be effected.

An analytical method suitable for mill use was developed to measure the amount of polyamide or polyurethane on a wool fabric and to distinguish between these two polymers. This method has special industrial interest because it offers a rapid and easy means to identify polyamide resin on the fabric and to determine the amount deposited in the WURLAN treatment of wool. It offers a suitable analytical quality control method for commercial WURLANizing.

2. Uniformity, Strength, and Nature of Yarns. The mechanism of lubrication of worsted yarns is being investigated by the Hosiery and Allied Trades Research Association in Nottingham, England under a P.L. 480 grant. Yarn is waxed and lubricated to impart an improved knitting behavior and appearance. In the early stages of this investigation it became obvious that a hitherto unsuspected factor obscured the true nature of the mechanisms involved in the waxing process. Evidence indicated that the natural oil and grease content on the yarn was higher than the amount of added wax and influenced the lubricity in an uncontrolled way and interyarn friction increased as wax pickup increased although the intent of waxing was to lessen such friction. Further work was conducted using deoiled yarn and the effect of waxing studied further. Wax melting point, temperature of waxing, yarn tension, and yarn speed affect the wax uptake by the yarn. Yarn flexural and torsional rigidities were found to be unrelated to wax pickup. A new apparatus was constructed to allow simultaneous waxing, measurement of friction, and knitting of yarn. Preliminary data indicate large differences between yarn to steel friction in knitting of waxed and unwaxed yarns with the wax reducing friction. Work will continue to obtain more significant data relating waxing to performance.

Studies on the uniformity and strength of yarns made from domestic wools of known history and selected fiber qualities were concluded. These studies were made to determine processing efficiency as related to fiber qualities. Spinning efficiency improvement by WURLANizing wool top was observed, making it desirable to study further of this approach to improving yarn strength before continuing basic studies on the effect of fiber qualities on spinning efficiency.

3. Fabric Construction. Both yarn and fabric construction greatly affect the performance of fabric and the response of fabric to chemical treatments to impart easy-care properties. The emphasis in research for the past year has been on the effects of the WURLAN treatment on mechanical processing characteristics. When WURLAN-treated wool top is spun, some of the deposited polymer tends to dust or flake off during gilling, drawing, and spinning, the amount depending upon the reagents and conditions used in the treatment. Oiling of the stock reduced dusting. More force is required to draft WURLAN-treated top or sliver than untreated top or sliver. Limited tests show that yarns from WURLAN-treated stock have higher tensile strength and slightly higher elongation at break than comparable yarns from untreated stock. No undue difficulties were found in spinning treated fibers, and present indications are that WURLAN treatment reduces yarn breakage during spinning.

4. Fluorochemical Treatment of Wool. Research has revealed a way to chemically graft a fluorine-containing resin to fiber surfaces. The new treatment makes wool oil and water repellent and resistant to shrinkage during machine laundering. The fabric has greater stability under acidic, alkaline, and bleaching conditions. By chemically anchoring the resin to the wool fiber, a durability to cleaning and abrasion was obtained. Cleaning and abrasion wore down all previous water and oil repellents.

5. Durable Creases in Wool Fabrics. Following extensive screening of chemical finishes for imparting durable creases to wool fabrics, ethanolamine in dilute aqueous solution was preferred. Research was concluded with this finding. Trials conducted by informal cooperation with the Quartermaster Research and Engineering Command proved the suitability of the ethanolamine treatment for military garments. Department scientists have provided specific processing directions to the defense supply agency in Philadelphia which has obtained equipment for treating garments with ethanolamine. The treatment will be written into the specifications for military garments as soon as the Quartermaster receives from the Surgeon General the final approval for the treated fabrics.

6. Bleaching of Wool. Research on procedures for bleaching wool is being conducted under contract by the Lowell Technological Institute in Lowell, Massachusetts. In present commercial bleaching treatments wool loses strength and resistance to alkali damage. The present contract investigation is defining limits for safely bleaching wool with hydrogen peroxide. Wool was bleached with peroxide in less than an hour at 120° to 160° F. at a pH of 9 to 10; however, the alkali solubility of the bleached wool increased at least 10%. Better reflectance without increased alkali solubility was obtained when the wool was reduced and cross-linked prior to or during the bleaching.

7. Improved Finishing Treatments for Wool Fabrics. The influence of different wool finishing procedures on fabric properties are being investigated by the Textile Research Association in Helsinki, Finland under a P.L. 480 research grant. Wool fabrics of three different structures were woven from three different grades of wool top. Half of each of the fabrics was woven from top-dyed yarn and the other half was piece-dyed. Progress indicates that the dyeing process can set the wool to desired smoothness similar to that obtained by special setting treatments. The best results with regard to surface smoothness after wetting and to evenness of dyeing were obtained when fabric was first open width scoured, then crab set or chemically set, and finally dyed in a strong acid dye bath. Only by thorough crabbing or chemical setting prior to rope dyeing could rope marks or weave distortion be avoided. The wool lost tensile strength when chemically set and dyed in a strongly acid bath. Neither scouring nor setting caused appreciable damage to undyed wool. Dyed pieces have not yet been tested. Experiments to devise a more effective setting process are being continued in attempts to produce, from coarser grades of wool, fabrics closer to those from finer grade wools. Coarser grades here refer to wools of 62's grade and coarser.

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Area 5 CITRUS AND SUBTROPICAL FRUITS-- PROCESSING AND PRODUCTS--WESTERN LABORATORY

Problem. The economic stability of the citrus and subtropical fruit industries in the Western Region is dependent upon effective utilization of fruit that cannot be accommodated on the fresh fruit market. The utilization of surplus or wholesome but blemished fruit provides the margin necessary to assure adequate returns to the farmer and continued development of stable markets. Ineffective utilization of products and continuously increasing processing costs are resulting in decreased returns to the growers. The California-Arizona grapefruit industry is encountering difficulty in disposing of both fresh fruit and processed grapefruit products. The pineapple and subtropical fruit industry in Hawaii must find practical methods for processing its products for export in order to prevent the accumulation of burdensome surpluses. The navel orange industry in California is hampered by the unavailability of satisfactory processes for the utilization of navel oranges. Juice extracted from early fruit, and during some seasons from almost all of the navel oranges, contains unknown substances that impart an intolerable bitter flavor to juice products after mild heat-processing or after standing at ambient temperature for a short time. Large new plantings of navel oranges may be expected to aggravate the utilization problem. Deterioration of the flavor and color of these and other processed citrus and subtropical fruit products imposes severe limitations upon the economic stability of the industry.

Information is needed on the chemical composition of citrus and subtropical fruits and their products and byproducts as a basis for the development or application of new and improved methods of processing; and for the production of new and improved food and industrial products and pharmaceuticals. Special attention needs to be given to the nature of the chemical changes involved during pre-treatment, processing and handling which lead to the formation of off-flavors, -colors, and -odors in processed products.

USDA PROGRAM

In the Western Utilization Research and Development Division, a concentrated program of fundamental research on citrus and subtropical fruit and its application to industry problems is conducted at the Division headquarters at Albany, California; at the Fruit and Vegetable Chemistry Laboratory in Pasadena, California; at the University of Hawaii, Honolulu; and, under a P.L. 480 grant, in Bogota, Colombia. Investigations are conducted on the composition of citrus essential oils, flavonoid compounds and other citrus constituents that are related to off-flavors and darkening of citrus products, the natural flavor components of oranges, enzyme systems that are involved in the appearance and disappearance of constituents and structures of plant tissues, constituents of dates that affect the quality and stability

of products, and the application of findings of such research to the development of new and improved citrus, tropical, and subtropical fruit products.

The Federal program of research in this area totals 18.0 professional man-years, including 3 scientists whose salaries are provided by two cooperators under Memoranda of Understanding (Lemon Products Technical Committee - 2, and the Date Administrative Committee - 1). Of the total 13.6 are assigned to investigations on chemical composition and physical properties; and 4.4 on new and improved food products and processing technology. In addition, the Division supervises a research project on development of new tropical fruit products supported by a P.L. 480 grant.

REPORT OF PROGRESS FOR USDA AND COOPERATIVE PROGRAMS

A. Chemical Composition and Physical Properties

1. Citrus Composition. The chemical composition and physical properties of citrus fruits determine the quality and stability of products made from them. Research is underway on the composition of cold-pressed lemon oil and lemon juice concentrate, the two principal lemon products in domestic and international markets, supported in part by the Lemon Products Technical Committee which pays the salaries of two research fellows at the Pasadena Laboratory. There are two problem areas. The first is concerned with differences between lemon juice and essential oil as they exist in the fruit and as they exist in products prepared for sale. These differences arise from the processes used in their manufacture and from handling the products in distribution. The second is the identification of components of lemon products over a wide range of growing areas to guide processors in improvement of processing methods. This information is also useful to lemon growers and processors to verify authenticity of lemon products in the market as a protection against unscrupulous competitors who market spurious products under the guise of authenticity, in violation of pure food laws. In both areas the approach has been to determine the composition of lemons and lemon products and to study the relationships of components to quality and to deteriorative changes that occur in the processing and subsequent handling of preserved products.

The terpenes (10-carbon hydrocarbon components) of lemon essential oil have been well characterized in earlier research and attention has been directed in the past year to the sesquiterpenes (15-carbon hydrocarbons). The principal sesquiterpenes of lemon oil have been isolated and the two most important (beta-bisabolene and bergamotene) identified. They have a mild odor and exist in sufficient quantity to add notes to lemon flavor. These two compounds make up about half of the sesquiterpene fraction, which is nearly 2% of whole lemon oil.

Studies of the carotenoids of lemon juice were initiated and 22 of these components, which are related to the color and nutritional qualities of lemon products, were revealed as natural components. These include

substantial amounts of phytofluene, zeta-carotene, and cryptoxanthin. Studies will continue on the further identification of carotenoids and will be extended to the steroid components.

Total amino acids, malic acid, total polyphenolic compounds, and titratable acidity were determined over a wide range of lemon juice samples known to be authentic. Limits of variability were established for several of these components to compare with analyses of lemon juice or concentrate of unknown purity. Characterization of individual amino acids will be continued. Methods for determining total carotenoids and steriods have recently been developed and are being tested.

2. Bitter Constituents of Citrus. Several classes of compounds that are natural constituents of oranges and grapefruit impart color and flavor to food products. During processing and in subsequent handling and storage, changes occur that affect the quality of the products. Clear understandings of the flavonoids in citrus and the quality contributions of various flavonoid compounds are a first step toward improvement of products and processes. Several flavonoids of citrus and citrus products have been identified and their contribution to flavor and color deterioration of preserved products has been determined. As reported in last year's report, naringin and neohesperidin are bitter constituents of the grapefruit and the bitter orange. There are also tasteless flavonoids in citrus. The flavonoids consist of a polyphenolic base connected to a compound sugar molecule. The bitter flavanones can be converted enzymatically to eliminate bitterness by separating the sugar from the base. They can also be debittered by another chemical reaction which has been accomplished in the test tube, converting the base to its equivalent dihydrochalcone. The bitter characteristic is related to a chemical linkage within the compound sugar molecule. Bitter flavonoids, when converted to the dihydrochalcone become very sweet. Tasteless flavonoids do not become sweet when converted to the dihydrochalcone.

A related flavonoid compound, phlorin, has a simple sugar rather than a disaccharide attached to the polyphenolic base. It is present in many citrus products and can participate in a browning reaction in the presence of amino acids which are also present in all citrus.

In these studies, new polyphenolic compounds were isolated and identified, including vitexin, in which the sugar is linked to the polyphenolic base by a carbon-to-carbon bond rather than carbon-to-oxygen-to-carbon linkage. Identification of these compounds and determination of their molecular structure involved analyses of components and degradation products, and synthesis of related compounds from precursors of known configuration.

Studies of limonoid compounds in citrus have been initiated. One, limonin, is an interesting, bitter compound found generally in citrus seeds and also found in the fruit of navel oranges, a seedless sport. Navel oranges

constitute a major part of new western plantings for the past several years. Attempts have been made to find an enzyme that will specifically change limonin to a tasteless compound. The only success in this work so far is with enzyme preparations that also settle the suspended material in the juice. Other methods of fragmenting limonin in order to debitter navel orange juice are under investigation.

The structural configurations of other highly oxygenated terpenoid bitter principles of citrus are also being studied in continuing research.

3. Fruit Flavor Components. Gas-liquid chromatography has rapidly advanced the chemistry of volatile components in a few years. Dual columns with dual-flame ionization detectors and programmed temperature control achieve remarkable sensitivity. The combination of gas-liquid chromatography with time-of-flight mass spectrometer separates and identifies substances in fractions of a part per million including materials whose separate existences are transitory. For the first time a class of flavor compounds called acetals have been detected in fruit volatiles. Some of the acetals have significant aromas and may contribute important flavor notes to fruits and fruit products.

Refinements in separation and detection of volatile components do not eliminate need for large-scale extraction to recover workable amounts of substances that exist in parts per million concentration in the original fruit. Identifications of some classes of flavor constituents continues to defy existing gas-liquid chromatography and other ultramicro methods. A solvent-extraction facility is nearing completion at Albany, California, that will allow for extraction with low flash-point solvents on a sufficiently large scale to make identifications not heretofore possible. The components of orange and other citrus juices will soon be studied on the same scale as investigations of commercially prepared cold pressed orange oil and other materials which have been obtained in large quantity under commercial operating conditions. Extractions will be made with these new facilities with much more knowledge of raw material than was ever possible from commercial samples.

Subjective evaluations of flavor are being made on purified organic compounds from food volatiles and believed to be related to flavor. A trained taste panel determines odor thresholds of aqueous solutions. This is one phase of a systematic approach to provide necessary correlations between the chemistry of volatile compounds and the flavor of foods. In order to study the effect of mixed systems, several compounds have been assembled in sub-threshold concentrations and evaluated together. From preliminary studies it appears that there is an additive relationship between chemical components in olfactory response. Thus, the aroma threshold of a ten-component mixture was identified by a test panel when each of the components existed at one-tenth of its individual threshold concentration. This finding is an early step toward interpretation in terms of flavor of the complicated chromatograms obtained in advanced chemical studies.

4. Composition of Dates. Utilization research is conducted with industry support by means of a fellowship salary provided by the Date Administrative Committee operating under a Federal Marketing Order. Objectives of the research are to study the natural components of dates, evaluate their relationships to quality of dates and date products, and to apply new information to the improvement of products and processes (see also paragraph B-2, Improved Date Products). Basic research, reported earlier, revealed compositional changes in dates that could be induced by naturally-occurring or added enzyme systems. Empirical studies guided by the research led to a commercial operation wherein dates were so treated that natural enzymes hydrolyzed sucrose to glucose and fructose and improved the flavor and texture. There appears to be more involved in this process than the simple inversion of sucrose, and further studies have been undertaken to elucidate other chemical actions involved in order to control commercial operations more positively.

Color deterioration of dates is now known to follow three pathways: (1) enzymic darkening, (2) oxidative browning, and (3) non-oxidative browning. Polyphenolic compounds that enter into the enzymic darkening of dates are being investigated. A caffeoylshikimic acid has been crystallized from date extracts and demonstrated to be a new enzymic browning substrate. This provides an important lead in the elucidation of date darkening. In order to obtain a more concentrated source of date flavonoids for chemical identifications, the date leaves were examined. Date leaves were found to have a high content of rutin, a compound with pharmaceutical properties. Because date cultural practices in this country are responsible for a large accumulation of leaves as a waste product, the presence of important components in leaves may offer another source of financial return for the grower. Investigations of date leaf components will continue along with the compositional studies of the dates.

5. Desert Grapefruit. Compositional studies on desert grapefruit and grapefruit products were recently initiated. Much work has been done on the composition of oranges and lemons in recent years using new methods of analysis and instrumentation. No equivalent compositional information has been obtained for grapefruit. A few scattered analyses are available on fruit of unknown cultural history, but most of these data were obtained using older methods of analysis, some of which are now considered inaccurate. Personnel have been assigned to this research and some routine analyses on minerals and vitamins will be handled by purchase contract. Separation and characterization of selected components of grapefruit will be carried out with emphasis being placed initially on sesquiterpenes, organic acids, carotenoids, sterols, and flavonoids.

6. Pharmacological Research. Caloric availability assays have determined the metabolic fate of citrus pectin ingested by rats receiving a restricted caloric intake. Pectin digestibility was low and, on the basis of weight gains, the small amount of pectin which was digested was not well utilized

as a source of energy for growth. Published findings outside the Department indicate that pectin included in the diet of rats at a rate of 5% will counteract the deposition of cholesterol and lipids in the liver which is induced by dietary cholesterol. The rats used in the pharmacological studies at Albany, California have been sacrificed and blood serum and liver samples have been frozen for later determination of cholesterol. The possibility that ingested pectin may inhibit the absorption of dietary or endogenous cholesterol from the digestive tract should be of interest to all concerned with blood cholesterol levels.

B. New and Improved Food Products and Processing Technology

1. Citrus Products. An important problem of the citrus industry is control of excessive bitterness in some products such as grapefruit and navel orange juice. There appear to be two major groups of compounds responsible for bitterness in citrus. These are the flavanone glycosides which cause the bitterness of grapefruit and Seville oranges and the limonoid compounds, a group of complex terpenoid substances that cause bitterness in navel orange products and in citrus seeds. As basic compositional research is conducted on these compounds, the new knowledge obtained is continuously applied in laboratory studies aimed at overcoming the flavor defects. Because the chemical structure of naringin, the bitter substance of grapefruit, has been elucidated it was possible to devise two ways to control this bitterness in laboratory tests. Enzymatic alteration of naringin by breaking the glucosidic link to form a non-bitter compound in grapefruit juice was accomplished in the laboratory but is not yet developed into commercial process. The slow reaction does not fit into large volume handling of the modern fruit juice plant. This bitter flavanone was converted to its equivalent dihydrochalcone in the laboratory. The new dihydrochalcone is intensely sweet, approximately 20 times as sweet as common synthetic sweeteners. Preliminary pharmacological tests indicate this new substance to be a safe food product. Several companies are now investigating the commercial applicability of this new product as a low calorie sweetener for foods. The market for low calorie sweeteners is currently about 4 million pounds per year and expected to expand to about 10 million pounds by 1970. The market now is being supplied by synthetic sweeteners produced from non-agricultural raw materials.

The debittering of navel orange juice by enzymic conversion of limonin is also being studied. No enzyme has yet been found that can make the chemical conversion without deleterious side effects such as clarification of the juice. Research directed toward finding a method for control of orange bitterness will be continued.

Knowledge of lemon juice composition is proving useful for determining purity of lemon juice products. In addition to their natural desirable flavors, lemon juice and concentrates contain large amounts of citric acid and are bought and sold on this basis for many food uses. Citric acid content is the most widely used measure of concentration of the juice. The lemon juice

producers were seeking a more exacting procedure to guarantee the authenticity of their product. Four quick, easy laboratory tests were used to establish a multiple correlation based on lemon juice of known purity. A formol titration for total amino acids, a polarimetric determination of L-malic acid, and a spectrophotometric estimation of the total polyphenolics were correlated statistically with titratable acidity, calculated as citric acid. The results of these tests were linked by computer, and limits were set to judge quality of unknown products in trade channels. The equation developed can describe authentic lemon juice by measurement of natural components that cannot be easily confounded by addition of inexpensive chemicals. An adulteration of as little as one-fifth of the natural citric acid content can be reliably detected. So small an addition would not reduce cost of product to a point where illegality is reasonably attractive. As a further refinement, measurements have been made on the individual amino acids, total carotenoids, and steroids of lemon juice. Preliminary results indicate that these substances are in proportion to citric acid. Their measurement may improve tests of lemon juice purity.

2. Date Products. As reported earlier, dates of softer texture and sweeter can be produced from hard and lightly flavored dates by treatment under conditions that accelerate the action of the enzyme invertase. Department studies leading to this new process were partially supported by industry funds. The process requires no new packing house equipment and has overcome a long standing problem of the domestic date industry. Normally about half of the United States crop fails to ripen to top quality and lacks soft, smooth texture. The new process involves heat activation of the natural date enzymes that convert sugar in dates to invert sugar. The conversion increases tenderness, improves flavor, and decreases the tendency of dates to dry out during storage. The proportion of dates harvested that will need to be processed by this improved method is increasing because of mechanical harvesting, in which all the dates on a particular tree are harvested at one time, precluding some of the natural ripening. The treated dates maintain their softness at low moisture levels and are especially suitable to new date and cereal products such as breakfast foods and baking mixes. Solution of the date texture problem has broadened domestic markets and places United States dates in a stronger competitive position for world markets.

3. Tropical Fruit Products. Agriculture in the new State of Hawaii has had severe economic problems. Caught between increased labor, package material, and freight costs on the one hand and foreign competition for traditional food markets on the other, it needs improved processing and new products to lower costs and improve quality so that the tropical and subtropical conditions in that State may be an adequate source of agricultural income. A new field station within the Western Utilization Research and Development Division has been established in Honolulu where cooperative research will be conducted with the Hawaii Agricultural Experiment Station. This research program will be focused on bulk and weight reduction of tropical fruits to yield high-quality, stable products for export to the

mainland or foreign countries. Improved means for retention or restoration of flavor in tropical fruit products will be investigated.

4. Foam-mat Drying. Laboratory and pilot plant investigations continue on the application of the foam-mat drying process invented by Department engineers at Albany, California. Cooperative research with the Southern Utilization Research Division is being conducted on the foam-mat drying of orange and other citrus products at Winter Haven, Florida. Informal cooperation continues with industry representatives who are interested in commercial application of foam-mat drying. Three commercial-scale configurations now exist. Two have been used for commercial-scale research. One drier has produced commercial lots of foam-mat dried tomato, pure lemon juice, and lime concentrate. Research has been conducted to further reduce the moisture of already foam-mat dried powders to eliminate the necessity for in-package desiccants for extremely hygroscopic materials. Rotating vacuum drying equipment, using lime as a moisture acceptor and an electrically heated outer cylinder has been designed. Orange powder can be reduced from 3-1/2 to 1% moisture by tumbling 2-1/2 to 3 hours at 130° F. A method has been developed to reduce the bulk and improve color of foam-mat dried powders. The powders are run between hot rolls which increase their density and provide for more efficient packaging. Conditions for rolling various products have been developed based on the sensitivity of the products to temperature and density specifications. For example, foam-mat dried orange powder is rolled at temperatures between 130 to 180° F. while tomato powder can be rolled at temperatures as high as 205°F. Orange solids have been prepared with a density of 0.8 grams per cubic centimeter which provides more solid material in a container than is present in a 60% frozen concentrate.

Important flavor components are frequently lost in the juicing, concentrating, and drying operations required to produce foam-mat dried or any other dehydrated fruit product. Methods are under investigation for adding back important flavor components in a stable form. The locking-in of orange oil with various types of sugars as carriers, has been used for sometime for citrus powders and formulated citrus flavored beverage powders. Recent research has provided locked-in orange oil at higher concentrations than previously available--up to 10%. The new equipment reduces the time the oil is in contact with melted sugars at a high temperature which might lead to deterioration of the oil. Work will continue to improve the quality of natural flavors which will be added back to foam-mat dried fruit products.

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Area 6 DECIDUOUS FRUIT AND TREE NUTS--PROCESSING AND PRODUCTS--WESTERN LABORATORY

Problem. Fruits and nuts are valued for their unique flavor, color, and natural vitamin content. In the period of abundance at harvest time, markets are glutted and growers often do not get an adequate return. Crops are perishable, and processing to preserve their unique qualities is difficult. No processed fruit retains completely the fresh values, although many highly acceptable products exist and about half of the fruits and nuts marketed in the United States are processed. Processing makes these commodities available to consumers the year around, and has opened new markets for producers. The proportion of processed commodities is steadily increasing but is dependent upon a continuing flow of new knowledge. Processing to preserve color, flavor, and texture presents many problems generally, and each new product requires the application of much scientific and technological skill.

The freezing process for preserving certain fruits keeps the products excellent at near fresh fruit condition. In spite of the gains in quality realized in freezing, many unsolved problems remain. The enzymatic browning of frozen peaches and the sloppy texture of frozen strawberries on thawing are two good examples.

Frozen fruits require expensive low-temperature storage and transportation facilities. The expense is greatly reduced by removing a portion of the water from the products. Orange and other fruit juice concentrates are well established in U.S. markets, and dehydrofrozen apple slices (rapid drying to 50% bulk weight and then freezing) are just becoming well established. Many other fruits and fruit juices should be amenable to concentration. Products of this type, however, are not so well adapted for export as those which do not require refrigeration.

The maximum weight reduction can be achieved through dehydration. The drying of fruit juices has been successfully accomplished by the vacuum puff drying and foam-mat drying processes. The latter is under intensive study, because it can be carried out at atmospheric pressure and consequently offers economy in processing. This process must be worked out in detail for many, as yet untried, fruit purees and juices and on pilot-plant scale for those products that show promise. Flavor recovery and the incorporation of recovered flavor in solid carriers for addition to the dried products require technological and basic chemical study. Essence recovery techniques developed for fruit juice concentrates are not completely satisfactory for this purpose.

Dried and canned fruits are now widely used in the U.S. and abroad. The popularity of dried fruits overseas and in this country would grow if stable, higher moisture dried fruits were available and if lower levels of sulfur

dioxide could be used without loss of quality. Control of mold spoilage in high-moisture dried fruit requires effective antimycotic agents.

Container costs for canned fruits limit the shipment of these products overseas. A solution of the container problem may be found in the use of lightweight fiber, foil, or plastic containers and aseptic filling procedures.

Fruit growers need new varieties of tree fruits and berries suited to processing and resistant to diseases endemic to each region of production. Utilization research is required in cooperation with farm research to assure growers of a market for fruit in the processing industry.

USDA PROGRAM

In the Western Utilization Research and Development Division, a broad program of basic and applied research on deciduous fruits and tree nuts is conducted at the Division headquarters at Albany, California; in field stations at Pasadena, California, Prosser and Puyallup, Washington; by contract in Honolulu, Hawaii; by grant at Cambridge, Massachusetts; and by grant funds under P.L. 480 in Israel. Fundamental research is conducted on fruit constituents that are involved in the flavor, color, and texture of fruit products, and includes development of laboratory tools to isolate and characterize individual components, investigation of such components as they occur naturally and as they are altered by operations involved in preservation, and the relationships between the components and the product values being preserved. Applied research is conducted to develop new and improved processes and products that will increase utilization of fruits and tree nuts, including the development of high quality concentrated and dehydrated products and more stable shelled tree nuts and the selection of improved processing varieties. Pioneering research on plant enzymes is also conducted.

The Federal program of research in this area totals 47.2 professional man-years, including two scientists whose salaries are provided by two cooperators (Dried Fruit Industry Research Advisory Committee, whose membership represents the California Raisin Advisory Board, the Dried Fig Advisory Board, the California Prune Advisory Board, and the Dried Fruit Association of California; and Diamond Walnut Growers, Inc. - one each), under Memoranda of Understanding; one contract providing research at the rate of approximately 0.5 professional man-years per year, and one grant providing research at the rate of approximately 0.5 professional man-years per year. Of this number, 24.5 are assigned to investigations on chemical composition and physical properties; and 22.7 on new and improved food products and processing technology. In addition, the Division sponsors basic research on fruit by means of a P.L. 480 grant.

REPORT OF PROGRESS FOR USDA AND COOPERATIVE PROGRAMS

A. Chemical Composition and Physical Properties

1. Fruit Pigments. Color preservation of many processed fruits involves the chemistry of the anthocyanin pigments which characteristically are red, purple, and blue. Investigations of natural and synthetic anthocyanidins continue to reveal new information concerning the structural relationships of individual compounds to color and the chemical changes that may be induced. An oxidative rearrangement of flavylium salts to 2-aryl-substituted benzofuran derivatives is of major scientific importance. It suggests the pathway both for color losses from fruit products and the natural occurrence of benzofurans, some of which are known to have biological activity. Because yields of benzofurans are very efficient (up to 50%) with starting materials that are easily obtained, this pathway for the synthesis of coumestrol and closely related compounds appears quite promising. Coumestrol, an important benzofuran, is a natural compound in alfalfa and legume forages which exhibits estrogenic activity. This newly discovered synthesis will become the basic approach to building a broad spectrum of estrogenic compounds to study their effects on animal metabolism in meat production.

Study of anthocyanins led to the elucidation of the role of sulfite in the bleaching of fruit products. Contrary to published views that decoloration resulted from the formation of a chalcone bisulfite addition compound, it has now been shown that chalcones derived from anthocyanins do not react with bisulfite. The decoloration is from a reaction of the anthocyanin to form a colorless sulfonic acid derivative. This explains the reversible nature of sulfite decolorizing. Synthetic flavylium compounds have been developed in a range of colors that offer promise of providing safe permanent color to improve processed fruits. Basic knowledge is being sought to block the chemical changes in anthocyanin pigments in fruit products so as to stabilize their color and thereby improve their quality.

2. Enzymatic Browning of Fruit. Basic research on enzymatic browning of fruit has led to a procedure whereby cut apple surfaces or peeled apples can be prevented from turning brown by surface adjustment of the acidity. A methyl-transferring enzyme with action similar to an enzyme from animal sources was postulated on the basis of the browning control of apples. Such an enzyme has now been found in two plant tissues. Apple cambium tissue and pampas grass shoots were two sources of the enzyme preparation, which has been characterized as a 3-O-methyltransferase. It has a high pH optimum and will permanently prevent the browning of natural catechol derivatives of fruit.

Related studies have been initiated under P.L. 480 at the Hebrew University in Jerusalem, Israel. Enzymes of several deciduous fruits are being investigated and identified by their substrate specificity and other properties.

Extraction methods and localization of phenolase enzymes in apples have been investigated and preliminary experiments made to follow changes in phenolase activity during the ripening of apples.

3. Chemical Origin of Plant Structural Tissue. Enzymatic pathways whereby polysaccharides and cellulose are formed and degraded in fruit are under investigation. The demonstration that inositol is a precursor of pectic material in the strawberry was reported. The continuing investigation reveals that biosynthesis of cell wall polysaccharides can stem from conversion of either D-glucose or myo-inositol into anhydropentose polymers. The pentose units derive from the 1 through 5 carbons of D-glucose and 2 through 6 carbons of myo-inositol. Preliminary information points to D-glucuronic acid as an intermediate in either case. A research grant at Harvard has recently been initiated to permit Dr. Porter to extend investigations in this area.

The chemistry of plant cell walls is under investigation because of the obvious importance of cell walls to such characteristics as texture, turgor, and cell elongation. In order to obtain uniform and reproducible cells, at least in the beginning of this work, in vitro tissue culture of tobacco cells was employed. The walls of these cells were found to contain a protein which was resistant to the usual protein extractants. It is also unique in the amount of the amino acids, proline and hydroxyproline. Characterization of this protein and its role in cell wall structure and properties will continue.

4. Fruit Flavor Components. Gas-liquid chromatography has rapidly advanced the chemistry of volatile components in a few years. Dual columns with dual-flame ionization detectors and programmed temperature control achieve remarkable sensitivity. The combination of gas-liquid chromatography with time-of-flight mass spectrometer separates and identifies substances in fractions of a part per million including materials whose separate existences are transitory. For the first time a class of flavor compounds called acetals have been detected in fruit volatiles. Some of the acetals have significant aromas and may contribute important flavor notes to fruits and fruit products.

Refinements in separation and detection of volatile components do not eliminate need for large-scale extraction so as to recover workable amounts of substances that exist in parts per million concentration range of the original fruit. Identifications of some classes of flavor constituents continue to defy existing gas-liquid chromatography and other ultramicro methods. A solvent extraction facility is under construction and nearing completion at Albany, California, that will allow for extraction with necessary low flash-point solvents on a sufficiently large scale to make possible identifications not heretofore possible. The components of deciduous fruits, berries, oranges, and citrus juices will soon be studied on the same scale as investigations on commercially prepared cold pressed orange

oil and other materials which have been obtained in large quantity from commercial installations. Extractions will be obtained in these new facilities with much more complete knowledge of raw material than was ever possible from commercial samples.

Subjective evaluations of flavor are being made on purified organic compounds from food volatiles and believed to be related to product flavor. A trained taste panel determines individual odor thresholds of aqueous solutions of these substances. This is one phase of a systematic approach to provide necessary correlations between the chemistry of volatile compounds and the flavor of foods. In order to study the effect of mixed systems, several compounds have been assembled in sub-threshold concentrations and evaluated together. From preliminary studies it appears that there is an additive relationship between chemical components and olfactory response. Thus, the aroma threshold of a ten-component mixture was identified by a test panel when each of the components existed at one-tenth of its individual threshold concentration. This finding is of great importance and an early step toward interpretation in terms of flavor of the complicated chromatograms obtained in our advanced chemical studies.

5. The Chemistry of Sulfur Dioxide in Dried Fruit. The color and nutrient qualities of several dried fruits are preserved by the addition of sulfur dioxide. In storage, as the sulfur dioxide disappears the fruit turns brown and nutritive qualities are lost. Chemical studies revealed that the disappearance of sulfur dioxide was caused by an oxidation of the compound to sulfate. In order to control this reaction the removal of oxygen from the package was indicated and an oxygen scavenger was devised based on the same oxidation that takes place in the sulfur dioxide conversion to sulfate. A reaction mixture of sodium bisulfite, activated carbon, ferric chloride, and hydrogen peroxide solution, was found to remove 97.5% of the oxygen from 280 milliliters of air in 24 hours. The reaction rate can be altered by varying the ratio between weight of the mixture and the air volume. Composition of the reaction mixture can be adjusted for the production of atmospheres at specific oxygen and carbon dioxide contents, such as are used in the storage of some fresh fruits and vegetables. Application of this new basic knowledge to the improvement of dried fruit products in laboratory experiments, is described in paragraph B-2 below.

6. Pharmacology of Dried Fruit Preservatives. Pharmacological studies, which have since 1960 been partially supported by the Dried Fruit Research Advisory Committee who provided the salary for a scientist hired to conduct the studies, have been concluded. Studies on methyl formate were initiated to obtain toxicity data to assure the safety of this compound. Work on methyl formate was discontinued when this compound was given FDA clearance. Toxicological studies on ethylene and propylene oxide and their derivatives were initiated. In view of the approval for use of propylene oxide on prunes and glacé fruits, work on this substance was not continued. Long-term feeding tests of diethylene glycol, the product of ethylene oxide and water, with laboratory animals were completed. These tests demonstrated

that diethylene glycol added to the diet increased urinary oxalate excretions only at feeding levels that were much higher than ever encountered in application of ethylene oxide to dried fruit. Bladder stones of oxalate were caused in rats, although bladder stones were not observed when diethylene glycol was fed at reasonable levels. Subacute toxicity studies for ethylene glycol in rat diets containing high-moisture dried fruit, indicated that the dried fruit (presumably because of its magnesium content) protected dogs and rats against oxalate bladder stones. Data obtained from this and from two-year feeding tests of diethylene glycol have been given to the Dried Fruit Association for transmittal to the Food and Drug Administration to support a petition for the use of ethylene oxide. In the final report of this research, evidence was presented supporting the conclusion that the commercial use of ethylene oxide as a sterilizing agent for dried fruits in the amount used and under conditions of use does not present a health hazard traceable to ethylene or diethylene glycols. Work is continuing to find a successful method for determining glycol residues in treated products. Such a method is required for clearance of ethylene oxide for use. Natural substances in dried fruit interfere with the determination of diethylene glycol. An effort is being made to avoid such complications and devise a suitable analytical method.

7. Microbial Flora in Fruits and Vegetables. Fundamental studies on the microbial flora within tissues of healthy fruits and vegetables have been concluded in Israel, where they were supported by a Public Law 480 research grant to the Agricultural Research Station of the Ministry of Agriculture at Rehovot. This study demonstrated the presence of certain non-pathogenic bacteria in normal healthy fruit and vegetable tissue. It was demonstrated that only certain types of bacteria are found within the tissue, that many types of organisms found on the surface are not found inside unless the fruit has been damaged, and that the flora found in fresh fruit remain harmless and have limited growth in the intact plant. The organisms do multiply during certain processing operations such as brining, where bacterial growth is necessary to the fermentation involved in preservation. In such cases conditions must be arranged so that the fermenting bacteria will overcome those present within the fruit and vegetables, otherwise an undesirable bacterial growth may bloat or putrefy the brined product.

8. Ethylene Metabolism in Fruit. The enzymes involved in the metabolic fate of ethylene in fruit under conditions wherein ethylene hastens the post-harvest maturation (i.e. accelerates the onset of the climacteric) are under investigation, using ethylene labeled both with C^{14} and with H^3. By measurement of radioactive compounds, it was shown that ethylene gives rise to a large number of volatile metabolites in the avocado. Approximately 12% of the total incorporated radioactivity of metabolized ethylene-H^3 was found in the methyl group of volatile toluene produced; and 5%, in the benzenoid portion of toluene. In addition, 1% of the metabolized ethylene was found in benzene. It was also demonstrated that 9% and 0.5% of the total incorporated radioactivity of metabolized ethylene-C^{14} were found in benzene and toluene, respectively. No one had foreseen

that ethylene would be metabolized to those hydrocarbons, or that such hydrocarbons could be found in mature, green avocados. Both discoveries contribute to our understanding of maturation. The isolation and identification of other volatile and non-volatile metabolites are continuing in order to obtain additional information about the enzymes involved in the maturation of fruit with the ultimate goal of controlling maturation prior to processing.

B. New and Improved Food Products and Processing Technology

1. Foam-mat Drying. Laboratory and pilot plant investigations continue on the application of the foam-mat drying process invented by Department engineers at Albany, California. Cooperative research with the Southern Utilization Research Division is being conducted on the foam-mat drying of orange and other citrus products at Winter Haven, Florida. Informal cooperation continues with industry representatives who are interested in commercial application of foam-mat drying. Three commercial-scale configurations of the foam-mat drying concept now exist. Two of these have been used for commercial-scale research. One drier has produced commercial lots of foam-mat dried tomato, pure lemon juice, and lime concentrate. Research has been conducted to further reduce the moisture content of already foam-mat dried powders to eliminate the necessity for in-package desiccants for extremely hygroscopic materials. Rotating vacuum drying equipment, using lime as a moisture acceptor and an electrically heated outer cylinder, has been designed. Orange powder can be reduced from 3½ to 1% moisture by tumbling 2½ to 3 hours at 130° F. A method has been developed to reduce the bulk and improve apparent color of foam-mat dried powders. The powders are run between hot rolls to increase their density and provide for more efficient packaging. Variations in rolling have been developed based on the sensitivity of the products to temperature and the requirements of density specifications. For example, foam-mat dried orange powder is rolled at temperatures between 130 and 180° F., while tomato powder can be rolled at temperatures as high as 205°. Orange solids have been prepared with a density of 0.8 grams per cubic centimeter providing more solid material in a container than is present in a 60% frozen concentrate.

Important flavoring components are frequently lost in the juicing, concentrating, and drying operations required to produce foam-mat dried or any other dehydrated fruit products. Methods are under investigation for adding back important flavor components in a stable form. The locking-in of orange oil with various types of sugars as carriers has been used for some time for citrus powders and formulated citrus beverage powders. Recent research has provided locked-in orange oil at higher concentrations than previously available--up to 10%. The new equipment reduces the time the oil is in contact with melted sugars at a high temperature which might damage the oil. Work will be continued to improve the quality of natural flavors to be added back to foam-mat dried fruit products.

2. Dried Fruit Products. Research on new and improved dried fruit products is continuing partly supported by the Dried Fruit Industry Research Advisory Committee, which provides the salary of a scientist assigned to this work. A process for making a non-setting raisin paste was developed. Raisin paste can be used in many bakery formulations. Heretofore it has set into a hard lump, so that bakers have had to prepare it just before use. A heat treatment was devised that softens raisin paste permanently. Department results were made available to the interested members of industry and successful pilot operations have been conducted. Factory-scale production of non-setting raisin paste is anticipated.

Stability of some types of dried fruit is dependent upon the protective action of sulfur dioxide. However, the preservative is not stable and as its concentration diminishes during processing and storage, so also does its protective action. Much greater conversion of sulfur dioxide occurs during sun drying than during controlled dehydration. Methods have been developed to maintain the level of sodium bisulfite. Low temperature storage and removal of oxygen from the package are two methods of improving sulfur dioxide retention, thereby stabilizing dried fruit quality. An oxygen scavenger system was devised that was highly effective in removing free oxygen from package headspace.

Metal sequestering agents have also been found to improve color stability of dried fruits by an inhibition of oxidation. The improved color stability in this case can be attributed to complexing of metals which would otherwise combine with natural polyphenols to form dark compounds.

Illumination of transparent packages had different effects for different dried fruits. Light shortened the storage life of apples markedly. Apricots were not visibly affected by illumination but lost about 20% more beta-carotene than controls stored in the dark. Dried peaches were essentially unaffected by illumination. The higher deterioration rate of illuminated dried apples implied a more rapid disappearance of sulfur dioxide.

3. Processing Quality of Northwest Soft Fruit and Berry Varieties. So large a proportion of red raspberries, strawberries, and cultivated blueberries produced in the Pacific Northwest are processed that new varieties must adapt to processing to succeed. Processing quality of varieties of soft fruit and berries under investigation by the Washington Experiment Station are being evaluated in cooperative research. During the past year, five varieties and 59 hybrid selections of strawberries were considered. Five established varieties and 11 new selections of raspberries, 20 varieties of blueberries, and 6 of blackberries were evaluated for processing quality. Processing quality, disease resistance, other yield factors, and harvest date, are combined with other horticultural characteristics to determine which varieties should be released for commercial use.

4. Improved Fruit Juices and Fruit Juice Processes. With expanding apple production in the Pacific Northwest, increasing consideration has been

given to the commercial concentration of apple juice. Research is under way on the influence of the degree of concentration on the gelling tendencies of the product. At 48% solids, gel formation was not observed; at 58%, gels occasionally formed; and at 65%, products frequently were found that formed gels. Studies of chemical and structural change in apple components are being conducted in order to overcome this limitation.

Commercial sales of clear red prune juice, based on product developments of the Department's Western Utilization Research Division, have been promising. Storage studies of this product revealed a color instability at high storage temperatures. The product and the puree from which it is made have been found to be very stable frozen. While the product would have a limited shelf life at 50° to 70° F., it is possible to freeze the juice or puree at harvest time and reprocess it any time later for distribution. Savings from the reduced inventory of individual packages (generally glass for this product) and from economic use of equipment in times of normal plant inactivity might compensate for the cost of freezing. This suggested procedure would be especially valuable if processing facilities are crowded at the harvest, which is generally the case in fruit canneries.

5. "Dry-Blanch-Dry" Fruit Process. Conventional sun-drying of fruits occasionally leads to product losses because of rain and high humidity or contamination during field exposure. The "Dry-Blanch-Dry" process for fruits was reported earlier. In contrast to other dehydration this new process imparts desirable translucence previously obtained only by sun-drying. Processing research has continued and conditions established for producing high-quality "Dry-Blanch-Dry" apricots, peaches, pears, raisins, and apples. The process made possible the drying of high-quality apple slices with lower sulfur dioxide content. Light, attractive golden raisins were prepared by the new process without sulfuring.

Exploratory studies on new approaches to dehydration of fruit pieces have been recently initiated.

6. Softening of Brined Cherries. Serious outbreaks of brined cherry softening have plagued the Northwest cherry industry from time to time in the past several years. Cooperative studies have been conducted with the Oregon and Washington Agricultural Experiment Stations and with interested processors and growers to determine the cause and to develop controls. Although the underlying reasons for softening of brined cherries have not yet been resolved, two treatments have been developed that appear to control the deterioration. Heating the cherries and brine to inactivate enzymes and addition of extra amounts of calcium chloride to the brine both prevent softening without adverse affect on the product texture. Brine disposal in cherry processing plants is a serious economic problem. Re-use of the brine from one year to the next would reduce cost of chemicals and avoid the disposal nuisance. Studies on the re-use of brines are being initiated.

7. Stabilizing Shelled Nuts. More and more shelled nuts are being marketed in transparent packages. These products are very attractive at first but they tend to darken and turn rancid rather quickly in the channels of trade. An investigation of what is involved in deterioration of shelled nuts is supported in part by the Diamond Walnut Growers, Inc., which supplies the salary of one chemist assigned to the Pasadena, California laboratory. Previous work at Pasadena resulted in a process that involved critical control of moisture content, use of antioxidants, and sealing in a package with an adequate moisture barrier and has had extensive commercial use. To provide further technological advances, a study has been undertaken of the components of walnuts and the way they vary as rancidity develops in storage. A large quantity of nuts has been extracted and separated into 43 dry fractions, most of which contain more than one compound. Further evaluation of these components will provide a base line for studies of nuts that have undergone deterioration. Kernels of walnuts from two progressive stages of rancidity have also been extracted. Qualitative and quantitative measurements are being made of changes in components. Preliminary results suggest that free amino acids or related compounds and volatile carbonyl compounds formed during oxidation of unsaturated fatty acids are involved in the rancidity.

Contract research on the stability of shelled Macadamia nuts is being conducted by the Hawaiian Experiment Station in Honolulu. Samples of three varieties of Macadamia nuts have been shelled and roasted and adjusted to three moisture levels. Storage tests are being conducted at different temperatures both in the dark and under standard lighting to determine the effects of processing and packaging on the stability of product. Preliminary chemical analyses and organoleptic evaluations of these samples have been conducted and the stability study will continue.

8. Grape Juice and Grape Products. Processing of Concord grape juice and the effects of agronomic practices on composition and quality of juice are under cooperative investigation in the Prosser, Washington, State Experiment Station. A five-year study of Concord grape concentrate, in which nearly 500 samples were analyzed and evaluated, has been completed. Agronomic and processing history is known for each of these samples and correlations will be run. The effect of storage conditions on the quality of Concord juice concentrate has been investigated. At 0° F. a greater loss of methyl anthranilate, an important flavor constituent, was observed than at 32° F. or 60° F. At 32° F. the concentrate was more stable than at 60° F. Grape concentrate and flavor essence that has been recaptured during concentration operations have different storage characteristics. It may follow that concentrate and volatile essences should be stored separately to be recombined when reprocessed shortly before shipping to market.

Research on viniferous grapes has been initiated in order to seek new products that might enlarge markets for surplus wine grapes. Principal attention has been directed towards Thompson seedless grapes. A concentrate of

Thompson seedless grape juice can be transformed to a bland, clear syrup by removing harsh flavor and dark color with activated carbon. The resulting product is a suitable syrup for fruit canning purposes and could bring an economic return for surplus grape juice concentrate. New methods for low temperature concentration are being investigated in order to preserve more flavor in grape products.

9. Fouling of Heat Transfer Surfaces. In the concentration of fruit juices and tomato products, efficiency lost through fouling of heat transfer surfaces results in serious economic loss. In earlier work, a resistance-impedance thermometer concept was developed to measure accurately surface temperatures and make related heat transfer measurements that indicated that fouling was markedly more rapid when products were being warmed than when they were being boiled, and further indicated that fouling was increased by increasing surface temperature, vapor fraction in evaporator tubes, and product viscosity. A relationship between protein denaturation and fouling has led to the initiation of studies on the effect of proteolytic enzymes on control of evaporator surface fouling. Measurements are being extended to rotating steam coil evaporators as they gain commercial acceptance. Simpler generalizations are being sought, so that information obtained in these studies can be applied to existing evaporator installations and to the design of new evaporators.

10. Canned Concentrated Peach and Apricot Puree. An investigation on the effects of temperature and temperature variations encountered during processing, transportation, and distribution of concentrated peach and apricot purees has been completed under a P.L. 480 grant to the Experiment Station for the Food Preserving Industries in Parma, Italy. The behavior of single strength and concentrated peach and apricot purees was studied during various stages of the concentrating and preserving processes and during storage at five temperatures, all above freezing. Objective methods for evaluating the quality of product were developed. Significant changes in organoleptic properties, amounts of tannin-like substances, vitamin content, and viscosity, caused by processing treatments were measured. It was concluded that the following methods can be used to evaluate qualitative changes in peach and apricot purees: (1) variation in thiamine or pantothenic acid content; (2) variation in reflectance color; (3) increase in volatile acidity; and (4) increase in hydroxymethyl furfural content. Although the changes that occur in peach and apricot purees during manufacture, concentration, and storage do not seriously affect the nutritive value of products, the acceptability of products is lowered during prolonged storage, particularly at higher temperatures. Storage temperatures lower than about 65° F., preferably below about 55° F., are needed to preserve organoleptic, physical, and nutritive properties of peach and apricot purees for long periods. This is particularly true for concentrates, since they are less stable than single strength purees under identical storage conditions.

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Area 7 POTATOES--PROCESSING AND PRODUCTS--WESTERN LABORATORY

Problem. The potato industry, faced with a continuing decline in the consumption of fresh potatoes, is becoming more and more dependent upon the development of new and improved processed products to maintain markets and to avoid recurring economic disasters. Crop perishability, supply fluctuations, and the inelasticity of demand, result in wide swings in price with even slight surpluses. In producing areas having a substantial processing industry, depressive lows are moderated by advance contracting by processors prior to harvest. However, in many important potato growing areas processing has not yet developed, and vulnerability not only still exists, but is exaggerated by the growing competition of processed potato and other competing food products. A continuing improvement in processed potato products is clearly required if processing is to expand fast enough to offset the progressive decline in use of fresh potatoes.

To improve the quality of processed potatoes, ways must be found to eliminate the stale, "earthy," rancid, "green," and "warmed-over" flavors that are sometimes encountered in potato products, including dehydrated mashed potatoes, dehydrated diced potatoes, frozen French fries, frozen patties, and potato chips. Equally important, methods must be devised to retain the desirable natural flavor of the freshly cooked potato in the processed product. Recently developed research methods offer an opportunity to isolate and identify the chemicals responsible for the natural flavors and the off-flavors, to develop rapid and sensitive analytical methods for their measurement, and to determine the raw material factors controlling formation of the various desirable and undesirable constituents in the fresh potato. Further improvement in the texture of potato products is also needed. Fundamental histological and chemical investigations could be used to determine the causes of differences in the texture of potatoes, as a basis for developing improved processing methods. Enzymes play a great part in the entire compositional pattern of the potato, not only the constituents responsible for flavor, off-flavor, color, and texture, but also those responsible for disorders such as "black spot." Black spot causes severe losses both to those who market potatoes fresh, and to those who process potatoes, because trimming costs are sharply increased and yields reduced. Increased knowledge of enzymes is needed as a basis for solution of the black spot and similar problems, to increase use of potatoes by reducing costs, and to improve quality of both fresh and processed potatoes.

USDA PROGRAM

In the Western Utilization Research and Development Division, basic and applied research on potato products is conducted at the Division headquarters at Albany, California, and by grant funds under P.L. 480 in

England and Sweden. The chemistry of potato flavor and the compounds involved in deterioration of potato products are studied to provide a basis for new and improved potato processes and products. Histochemical studies are conducted to elucidate factors involved in the texture of potato products. Basic investigations on the enzyme systems involved in potato product discoloration and the role of sulfur dioxide in preventing non-enzymatic browning are in progress.

The Federal program of research in this area totals 9.7 professional man-years. Of this number, 5.5 are assigned to chemical composition related to flavor, color, and texture of potato products and 4.2 to technological and engineering research on new products and processing methods. In addition, the Division sponsors two research grants under P.L. 480 on basic studies.

REPORT OF PROGRESS FOR USDA AND COOPERATIVE PROGRAMS

A. Chemical Composition Related to Flavor, Color, and Texture of Potato Products

1. Flavor Components. The chemistry of volatile components connected with food flavor has advanced spectacularly in recent years by application of gas-liquid chromatography. Dual columns with dual-flame ionization detectors and programmed temperature control achieve remarkable sensitivity. The combination of gas-liquid chromatography with time-of-flight mass spectrometer separates and identifies substances in fractions of a part per million including materials whose separate existences are transitory. Research in this area has been supported in part by the Instant Potato Granule Manufacturers Association, who have supplied the salary of a scientist assigned to studies on volatile constituents of potatoes. (The position is currently vacant.)

Refinements in separation and detection of volatile components do not, however, eliminate need for large-scale extractions so as to recover workable amounts of substances that exist in the parts per million concentration range of the original food. Identifications of some classes of flavor constituents continue to defy existing gas-liquid chromatography and other ultramicro methods. A solvent extraction facility is under construction and nearing completion at Albany, California, that will allow for extraction with necessary low flash point solvents on a sufficiently large scale to make progress on identifications not heretofore possible.

Studies have continued on the volatile constituents of potatoes that are involved in flavor. When cooked potatoes are held in a wet mashed state a very rapid oxidation of lipids occurs. The volatile compounds produced in this oxidation seem to be largely lost in dehydration. However, non-volatile peroxides remain and may trigger subsequent autoxidation in dehydrated products. Small concentrations of antioxidants and chelating agents during the mashing operation prevent subsequent autoxidation. This laboratory work indicates that potato granules may be more stable if antioxidants

or chelating agents are added directly after the cooking step. Preliminary studies on storage of commercially prepared potato granules containing five parts per million of the antioxidant BHT indicate that a close correlation exists between the amount of hexanal formed and the off-flavor development.

In the gas-liquid chromatography of volatiles above potatoes and potato products a number of aldehydes were separated and identified. As a step toward demonstrating the contribution of each substance to the overall flavor development, a number of these aldehydes were synthesized in the laboratory for comparison with identified compounds from the potato products. Closely related compounds were also synthesized and evaluated to compare flavor with the natural compounds. The synthetic approach is providing basic information that will help lead to identification of compounds now separated from the volatiles of potato products but not absolutely identified.

Subjective evaluations of flavor are being made on purified organic compounds that are found in food volatiles and believed to be related to product flavor. A trained test panel determines odor thresholds of aqueous solutions of these substances. This is one phase of the systematic approach to provide correlations between the chemistry of volatile compounds and the flavors of food. In order to study the effect of mixed systems, several compounds have been put together in sub-threshold concentrations and evaluated. Preliminary studies indicate an additive relationship between chemical components and olfactory response. Thus, the aroma threshold of a ten-component mixture was identified by a test panel when each of the components existed at one-tenth of its individual threshold concentration. This finding is an early step toward interpreting in terms of flavor the complicated chromatograms obtained in advanced chemical studies.

2. Enzymic Browning. Fundamental studies on the enzymic browning of potatoes are in progress at the Low Temperature Research Station at Cambridge, England, supported by a grant under Public Law 480. This research is aimed at determining the nature, distribution, and mode of action of enzymes responsible for enzymic browning of potatoes and potato products and, further, determining the nature and distribution of phenolic substrates and pathways of their synthesis. During maturation of potato tubers, they pass through a phase in which there is unequal browning of the cut surface. In these immature tubers the browning is confined almost entirely within the vascular ring, the tissue outside the vascular ring browns only slightly. Analyses were made of the concentration of tyrosine, the concentration of the phenolase enzyme, and the susceptibility to enzymic discoloration across the cut surface of such tubers. Data obtained confirmed earlier work that indicated a close correlation between the tyrosine content and the degree of enzymic discoloration, but a less close correlation between the concentration of the enzyme phenolase and browning. Further, the experiment confirmed that the concentration of the other major phenolic constituent, chlorogenic acid, little influences the susceptibility of the cut surface to discoloration because of its light-colored oxidation product. A simple

experiment showed that if another enzyme substrate is applied across the entire tuber surface, then the browning is as rapid in the tissue outside the vascular ring as inside it, confirming that concentration of the enzyme is not limiting the rate of browning. The differential distribution of tyrosine disappears before the tuber reaches maturity, so that browning is then uniform over the cut surface.

With this knowledge in hand it is possible that effect of climatic, cultural, or kinetic factors on enzymatic browning may be studied in terms of the influence of these factors on the synthesis of tyrosine in the growing tuber. Greenhouse studies indicate certain fertilizer enrichment programs that can increase the concentration of tyrosine as much as three times. Fundamental studies of the biosynthesis of phenolic compounds and enzymes related to their formation are in progress.

3. The Role of Sulfur Dioxide in Dehydrated Vegetables. A basic investigation to determine the chemical mechanism of the protective action of sulfur dioxide to control browning in dehydrated vegetables is being conducted at the Covent Garden Laboratory in London, England, supported by a grant under P.L. 480. As this program approaches completion, excellent progress is shown by publication of eight scientific papers already, and with more on the way. Without adequate protection, many dehydrated fruits and vegetables gradually become brown, develop objectionable flavors and odors, and suffer losses in nutritive value. While the most common way to avoid these effects has been to treat the products with sulfur dioxide during processing, until recently very little has been known about the mechanism of this treatment. It has long been known that chemical browning reaction occurs between sugars and amino acids. This chemical reaction is being greatly clarified. When sugar solutions are heated, configurational changes occur, developing unsaturation in the sugar molecule. The rate of browning of such solutions depends on the ease of dehydration of the sugar-derived compounds and upon the reactivity of intermediates produced. These factors depend upon the molecular configuration of the sugar present initially and on the environment. Where sulfite is added to prevent discoloration of sugar and amino compounds, it has to deal not only with the initial sugars but also with the new compounds formed. Sulfite reaction with the initial sugar hinders the formation of some of these new compounds whose reactivity in terms of discoloration is greater than the initial molecular configuration of the sugar present. This new knowledge provides a basis for developing new methods for protecting dehydrated fruits and vegetables from nonenzymic browning, where sulfur dioxide is not satisfactory or its use must be limited.

B. Technological and Engineering Research on New Products and Processing Methods

1. Dehydrated Potato Granules. In the production of instant mashed potato powder, a major problem exists in the translation of laboratory results to plant scale, especially in the reduction of potato from a wet mash to a

coarse moist powder. The moist powder must be separated into individual cells or small cell clusters without undue rupture of cell walls and must not reagglomerate before or during subsequent finishing drying. Although many processes have been reported during the past 40 years, only processes involving add-back of previously dried materials have been successful for commercial production of high density granules. Research over the past several years has been conducted to develop a direct process involving three phases of drying: (1) an initial partial drying to about 55 to 60% moisture content; (2) an intermediate drying operation combined with simultaneous mixing of the product, and (3) a finish drying step. The initial drying has been performed by either of two techniques; drum drying of cooked mash, or through-flow conveyor drying of cooked half-inch cubes. In order to progress to the moist coarse powder stage a freezing and thawing operation has been used between the first two drying phases. After thawing, the intermediate drying operation has been performed in a specially designed trough-type dryer in which the product is continuously and gently mixed. New equipment has been designed in this investigation, including a continuous potato ricer, a through-type granulator-dryer, and granulating rolls. Descriptions of this new equipment have been made available for commercial potato processors to evaluate in connection with their potato drying processes.

PUBLICATIONS AND PATENT REPORTING RESULTS
OF USDA AND COOPERATIVE RESEARCH

Chemical Composition Related to Flavor, Color, and Texture of Potato Products

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Technological and Engineering Research on New Products and Processing Methods

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Area 8 VEGETABLES--PROCESSING AND PRODUCTS--WESTERN LABORATORY

Problem. Vegetable crops, in general, are perishable and seasonal and thus, are subject to supply and price fluctuations to the disadvantage of the agricultural economy. In order to expand markets and stabilize prices, new and improved processed products are needed that will be more desirable to the domestic and foreign consumer from the standpoint of quality, convenience, stability, nutritive value, safety, and cost. The quality of processed vegetables and the economy of their processing have not improved rapidly enough to increase or even maintain the relative position of vegetables in the American diet, or to increase substantially their contribution to the export trade. The consumption of dry beans and certain other vegetables is limited by the fact that they cause flatulence.

New easy-to-prepare vegetable products are needed, particularly from such commodities as dry beans and peas, which now require hours to prepare. The severe heating required to sterilize low-acid foods, which include most vegetables, seriously impairs the quality of canned products. The stability of all kinds of processed vegetables needs to be improved so that quality and nutritive value will be better preserved during storage and distribution. The safety and effectiveness of new chemical additives, needed to improve the quality and stability of processed vegetables, must be established. Better methods of removing residues of agricultural chemicals from vegetables for processing are urgently needed, as are procedures for decontaminating vegetables exposed to radioactive fallout. Of vital importance is research to reduce the costs of processing in order that the farmer may receive a larger share of the consumer's dollar.

Applied research on these practical problems must be supported by a strong program of basic research on the chemical constituents of vegetables responsible for flavor, color, and texture; on the reactions these compounds undergo before, during, and after processing; on constituents having biological activity; on the microscopic structure of vegetables and vegetable products; and on the micro-organisms which cause spoilage or loss of quality in these products.

USDA PROGRAM

In the Western Utilization Research and Development Division, a broad program of basic research on vegetables and the application of science to new and improved products and processes is conducted at the Division headquarters at Albany, California, in field stations at Pasadena, California and Puyallup, Washington; by contract at Urbana, Illinois, and Davis, California; and by grant funds under P.L. 480 in Cambridge and Chipping-Campden, England. Fundamental studies are conducted on the chemistry of

vegetable flavor and vegetable pigments, the mechanism of heat resistance in bacterial spores, the composition of dry beans as related to cooking quality and flatulence-producing characteristics, the factors affecting deterioration of dehydrated vegetables, and the microbiology of raw vegetables for processing. Applied research is conducted to develop new and improved products to increase the utilization of vegetables including new, high quality concentrated and dehydrated products and products of improved convenience of use, processes for producing these, and selection of improved processing varieties.

The Federal program of research in this area includes a total of 43.5 professional man-years including two scientists whose salaries are provided by the California Lima Bean Advisory Board operating under a State Marketing Order; two, by the United States Brewers Association; one, by the National Association of Frozen Food Packers, and one on half-time, supported by the National Canners Association. Of the total, 26.8 are assigned to chemical composition and physical properties and 16.7 to new and improved food products and processing technology. In addition the Division sponsors three grants under P.L. 480 on basic research.

REPORT OF PROGRESS FOR USDA AND COOPERATIVE PROGRAMS

A. Chemical Composition and Physical Properties

1. Vegetable Flavor Components. Flavor investigations of vegetables have been advanced by continued use of highly sensitive new equipment for the separation and identification of volatile components. Sulfur-containing compounds known to be powerful odor producers are being identified in green peas and hop oil. Basic studies on the sulfur compounds related to flavor of onions, cabbage, and related vegetables are approaching conclusion. A new basic study of flavor constituents of tomatoes has been initiated. Research emphasis has been placed on the sulfur-containing constituents and other volatile compounds of vegetables in order to develop objective methods for estimating flavor of vegetables to control flavor loss or deterioration resulting from chemical or enzymic action, and restore the flavor in dehydrated products. Volatile sulfur-containing components from onions have been extracted, separated, and identified. A new colorimetric reagent, N-ethylmaleimide, was discovered which will distinguish thiolsulfinates from the closely related thiolsulfonates, when both are involved in the flavor of onions. The amino acids that are precursors to the volatile sulfur-containing compounds were identified in onions. An enzyme preparation from onions that is capable of transforming the flavor precursors present to volatile flavors, was isolated and characterized. This enzymic reaction was identified, and pyruvic acid determined to be an important reaction product. It was demonstrated that the flavor of dehydrated onions can be enhanced by drying under conditions that will preserve this enzyme system. The production of pyruvate from this enzymic reaction formed the basis of a test for onion pungency. The quantity of pyruvate produced in the enzymic reaction is a measure of the pungency of either fresh or dehydrated onions.

Values obtained by the simple and rapid procedure were found to correlate well with taste panel determinations of the olfactory threshold of onions. The test is useful and has been adopted by the domestic dehydration industry for selecting raw materials for processing and by plant breeders to obtain varieties of onions more suitable for processing.

The chemistry of volatile components connected with food flavor has advanced spectacularly in recent years by applying gas-liquid chromatography techniques. High instrument sensitivity is achieved using dual columns with dual flame ionization detectors and programmed temperature control. A new combination for separation and detection of volatiles was instituted using gas-liquid chromatography in connection with the time-of-flight mass spectrometer. This combination provides separation and identification of incredibly minute amounts of substances, including materials whose separate existences are transitory. Refinements in separation and detection of volatile components do not eliminate need for large-scale extraction so as to recover workable amounts of substances that exist in the parts per million concentration range of the original food product. Identifications of some classes of flavor constituents continue to defy existing gas-liquid chromatography and other ultramicro methods. A solvent extraction facility is under construction and nearing completion at Albany, California that will allow for extraction with necessary low flash point solvents on a sufficiently large scale to make possible identifications not heretofore possible. Extractions will be obtained in these new facilities with much more complete knowledge of raw material than was ever possible when working with commercial samples.

A basic study of the volatile flavor components of hop oil is supported in part by the United States Brewers' Association, which provides the salaries of two chemists. The long-range purpose of this project is to isolate and characterize flavor components and to study their chemistry. Analysis of one sample of hop oil provided mass spectra for 59 compounds, 23 of which were identified by their mass spectral patterns and gas-liquid chromatography retention times, 11 others were tentatively identified, and empirical formulas were obtained for most of the other compounds. Analyses have been obtained on hop oils from five different varieties using temperature programmed, capillary gas-liquid chromatography and by chemical separation methods. Samples of hops that had been evaluated for aroma by the United States Brewers' Association Hop Committee were obtained to compare direct vapor injection gas chromatograms with the aroma results. Preliminary findings indicate a correlation between the Hop Committee's aroma evaluation and the gas-liquid chromatography pattern obtained.

Cooperative work, supported in part by the National Canners Association, which provides the salary of a chemist assigned half-time to the project, is conducted on the composition of peas, to provide a basis for technological improvement of canned pea flavor. This project has attempted to develop instruments to measure flavor more objectively and gather more complete data on the composition of volatile components from peas. Considerable progress

has been made in past years in identifying components of peas, many of which exist only in minute concentration. To obtain suitable quantities of volatile materials from peas for chemical studies, volatile material was collected above a commercial pea blancher. The collection equipment was operated continuously for 17 days and a total of 200 gallons of condensate was collected. Vapor above the condensate was collected on activated charcoal in a trap. The volatile material from peas was extracted from the charcoal with various solvents. Separations and identifications were made using gas-liquid chromatography and other procedures. Studies were initiated to seek correlations between individual compounds detected by these sensitive methods and organoleptic evaluations.

Subjective evaluations of flavor are being made also on purified organic compounds, such as those from volatiles which we suspect affect product flavor. A trained panel determined individual odor thresholds of aqueous solutions of these substances. This is one phase of a systematic approach to provide necessary correlations between the chemistry of volatile compounds and the flavor of foods. In order to study the effect of mixed systems, several compounds have been assembled in sub-threshold concentrations and evaluated. From preliminary studies it appears that there is an additive relationship between chemical compounds in olfactory response. Thus, the aroma threshold of a ten-component mixture was identified by test panel when each of the components existed at one-tenth of its individual threshold concentration. This finding is an important early step toward interpretation in terms of flavor of the complicated chromatograms obtained in advanced chemical studies.

2. Nature of the Heat Resistance of Bacterial Spores. The extreme resistance of bacterial spores to heat necessitates processing treatments which bring about flavor, color, texture, and nutritional deterioration in canned vegetables. A basic investigation of the nature of heat resistance exhibited by spores is expected to uncover facts that will make milder processes possible. The temperature dependent heat adaptation of B. megaterium spores has been studied in the temperature range of 4 to 65° C. Heat resistance increases of 50-fold were observed. The uptake of cations by spores in neutral calcium salt solutions indicates that heat adaptation consists of calcium loading of the spore cation binding system. This new knowledge of the high cation exchange capacity of dormant spores and structural information obtained by electron microscopy tend to support, although not prove, the hypothesis advanced by Department scientists that spore resistance to heat is connected with a dehydration process of constriction in the dormant spore rather than with a theoretical water-impermeable barrier. Fractionation and identification of spore components and structures have been materially advanced by the dry rupture of lyophilized spores in the presence of a large excess of sodium chloride crystals and by studies of ash residue following ultramicro incineration at room temperature with activated oxygen. These new techniques should be helpful in arriving at an ultimate understanding of the heat resistance of bacterial spores.

3. Composition of Dry Beans. The composition of dry beans is being investigated to determine the factor or factors involved in their intestinal effects. A simple, portable apparatus was developed for measuring the amount and composition of human flatus from subjects who have eaten experimental meals of cooked beans. The apparatus is worn by human subjects without undue interference with their normal activities. Flatus is collected and passed through three small glass tubes, one of which contains chemicals that capture the passed carbon dioxide which is one of the major gaseous components of human flatus. Periodic samples of gas are measured by gas-liquid chromatography to determine hydrogen, methane, oxygen, and nitrogen as well as the carbon dioxide. The total carbon dioxide collected and the composition of gas taken at intervals allows for calculations of the total flatus and its composition over a many-hour period following ingestion of cooked beans. Using this human assay technique, a bean fraction has been isolated which contains the major portion of flatulence activities of the whole bean, but represents less than 8% of the original whole cooked bean. Preliminary investigations indicate that this chemical fraction is made up of compounds of molecular weight less than 10,000. This fraction contains a great number of chemical compounds, including peptides, pigments, tannins, free amino acids, monosaccharides, and protein complexes involving lipids and carbohydrates. It probably does not contain higher molecular weight materials such as proteins, pectins, gums and dextrans, cellulose and starch.

By informal cooperation with the Clinical Investigation Center of the United States Naval Hospital in Oakland, California, medical supervision is given to these studies. In addition, because the flatulence principle in beans is thought to increase peristalsis in the human digestive tract, the Naval Hospital has undertaken a cooperative study of peristalsis. Two procedures are being used to study peristalsis. In one, a small radio transmitter that responds to minor pressure differences is swallowed and peristaltic action, as reflected by pressure differences in the intestines, can be recorded. The differences in the pressure patterns following normal diets and diets containing controlled amounts of beans, and the transit time of the transmitter through the intestines, is providing useful information on determining the effect of bean diets on intestinal reaction. In addition, the transit time for gas passage through the intestines is being measured by administering oxygen by stomach tube to a subject after a normal diet and bean diet, and noting any difference in the time required for an increased oxygen-to-nitrogen ratio to appear in the flatus.

From time to time, commercial samples of bean products and recipes and additives suggested to prevent flatus have been obtained and tested by the new assay technique. To date none of these samples or ideas has been found to reduce the flatulence which follows ingestion of dried beans.

Closely coordinated with these studies at Albany, California is supporting research conducted by contract at the University of Illinois. Methods for measuring flatulence in human and animal subjects are being developed, and research is underway on the physiological mechanism underlying flatulence.

A simple portable electronic device for measuring and analyzing egested flatus has been devised and is being tested. If successful, this technique will be particularly useful when experiments are to be conducted with large panels of human subjects. In addition, test tube studies have been conducted in which cooked dry beans have been found to catalyze the release of carbon dioxide gas from solutions containing bicarbonate ions. The pancreas normally secretes large amounts of bicarbonates into the upper intestine.

Animal studies are continuing at Albany, California both feeding beans to rats and by injecting bean components in their upper intestines when anesthetized. As previously reported, irritation of intestine walls and intestinal swelling have been observed. Research procedures have been refined so that sections of the intestine can be excised and weighed. It has been found that weights of treated strips of intestines are 50 to 70% heavier than comparable controls when an injection of ethanol extract of beans has been made into an intestinal loop. Earlier tests were made with pentobarbital anesthesia which restricts peristalsis. Urethane now allows peristalsis to continue in these studies. The injection of the alcoholic fraction results in increased peristalsis and distension of the intestine with gas as well as with mucus. The largest volume of gas was produced when bean extracts were inserted into the rat's stomach by stomach tube. Starved rats are used for this experiment and peptone was added to stimulate the flow of gastric juices. The effect of the bean extract was striking.

4. Composition of Beans and Peas as Related to Cookability. Compositional studies, supported in part by the California Lima Bean Advisory Board, which provides the salary of two scientists, are directed towards understanding the effect on cooking of qualitative and quantitative changes in the proteins of large Lima beans. Changes in cooking characteristics of Lima beans were found to parallel closely changes among the proteins, which comprise about 25% of dry bean solids. Basic information will be required in continuing work to determine the specific chemical and physical changes among proteins during maturation, processing, and storage which may change the cookability of beans. In storage studies with large Lima beans, pintos, and Sanilacs (a Michigan pea bean variety), moisture content of beans has a dramatic effect on the maintenance of their cooking qualities during storage. Minimum changes in cookability occurred in low-moisture beans (between 8 and 10%) while moderately high-moisture beans (12 to 14% required 5 to 10 times longer cooking after a year's storage at 90° F. and several-fold more cooking time after storage at 70° F.

A basic investigation of compositional factors that might be related to the cookability of dry peas is being conducted at the Fruit and Vegetable Canning and Quick-Freezing Research Association Laboratories at Chipping-Campden, England, supported by a grant under P.L. 480. It was reported earlier that the texture of cooked dry peas is determined by the viscosity of the intercellular pectic material and the extent to which tissue cells swell during cooking.

The extensibility of cell wall was shown to dominate texture. Phytic acid concentration, which was earlier thought responsible for texture, was shown to be of little or no significance. In test tube studies, insoluble salts of phytic acid were formed in the presence of potassium, calcium, and magnesium ions, the composition of the insoluble salts depending upon the ratio of the cations present. When peas were cooked in water containing calcium ions, only part of the absorbed calcium was complexed as an insoluble salt of phytic acid. As the external calcium concentration was raised an increasing amount of calcium was absorbed at other sites in the tissues, which it toughened. As the insoluble phytic acid salt increased, a greater proportion of the magnesium which is naturally present in the peas was complexed in the salt, leading to a replacement of magnesium by calcium in other parts of the tissue. Histological studies showed that in the uncooked pea, calcium was predominantly in the inner cell wall. Cooking drives this calcium into the cell contents. As the cell wall loses calcium its plasticity is increased, allowing greater cell expansion and a corresponding softening of the tissue. When peas are cooked in hard water, or in the presence of calcium salts, the removal of calcium from the cell wall is counteracted by the absorption of calcium from solution and the peas remain tough.

5. Vegetable Pigments. The retention of natural color in processed vegetables affects the acceptability of products. Basic research is conducted at the Department's Laboratory in Albany, California on chlorophyll and at the Low Temperature Research Station in Cambridge, England under a P.L. 480 grant on carotene and its volatile oxidation products. Research in England has shown that color of processed vegetables is related tc other quality factors. Beta-carotene, a precursor of vitamin A, is the principal yellow color in carrots. As this color disappears, there is a concurrent development of undesirable odors and flavors which are breakdown products of the carotenes. Investigations of the complicated chemical mechanisms involved in the breakdown of beta-carotene are being conducted in the expectation that means will be found to stabilize vitamin A, and improve the color and nutritional quality of processed yellow vegetables, and avoid undesirable flavors that may result from processing and subsequent storage of processed products. Chlorophyll derivatives and degradation products are being measured by various laboratory procedures. Column chromatography, radial and centrifugal paper chromatography, and thin-layer chromatography are being employed. Highly purified chlorophyll a, chlorophyll b, pheophytin a, and pheophytin b, have been obtained as reference standards for analytical work. Separation of pheophytin a from pheophorbide a needs further refinement. The extreme instability of chlorophyll immensely complicates the research in this area.

6. The Role of Sulfur Dioxide in Dehydrated Vegetables. A basic investigation to determine the chemical mechanism of the action of sulfur dioxide to control browning in dehydrated vegetables is being conducted at the Covent Garden Laboratory in London, England, supported by a grant under P.L. 480. As this program approaches completion, evidence of excellent progress is noted in the publication of eight scientific papers with more anticipated.

Without adequate protection, many dehydrated fruits and vegetables will gradually become brown, develop objectionable flavors and odors, and lose in nutritive value. While sulfur dioxide has stopped browning, until recently very little has been known about the mechanism of this treatment. Chemical browning reaction occurs between sugars and amino acids. This chemical reaction is being greatly clarified. When sugar solutions are heated, configurational changes occur, developing unsaturation in the sugar molecule. The rate of browning of such solutions depends on the ease of dehydration of the sugar-derived compounds and upon the reactivity of intermediates produced. These factors depend in turn upon the molecular configuration of the sugar present initially and on the environment. Where sulfite is added to prevent discoloration of sugar and amino compounds, it has to deal not only with the initial sugars but also with the new compounds formed. Sulfite reaction with the initial sugar hinders the formation of some new compounds whose reactivity in terms of discoloration is greater than the initial molecular configuration of the sugar present. This new knowledge provides a basis for developing new methods for protecting dehydrated fruits and vegetables from non-enzymic browning, where sulfur dioxide is not satisfactory or its use must be limited.

7. Histological Studies of Vegetable Tissues. Basic studies on the texture of frozen vegetables were initiated with support from the National Association of Frozen Food Packers, which supplies the salary of one scientist. Shear press texture evaluations were made of green beans following blanching, freezing and thawing, and cooking operations. A substantial shear press reading decrease following freezing and thawing indicates this as a major area in which to concentrate investigations of textural change. Two types of damage were observed microscopically in frozen tissue: the thin-walled parenchyma cells were broken, and thick-walled cells were separated. When freezing was very rapid, as by immersion in liquid nitrogen, these two types of damage were not observed. To determine whether the freezing or the thawing operations were principally concerned with the textural changes, two techniques were used to observe the effect of freezing without thawing. In one, frozen green beans were dried under vacuum so as not to thaw and to maintain general tissue configurations and, secondly, the ice was removed from frozen tissues at very low temperature by alcohol. If the freezing is done slowly, damage seems to occur during the freezing so that there is no substantial additional change during thawing. If the product is frozen more rapidly there is little damage during freezing but damage resembling that of the freezing can occur during a subsequent slow thawing operation. Neither blanching nor cooking caused the changes of the freezing and thawing operations. Studies will continue to acquire a better understanding of the physical and chemical changes during freezing and thawing that affect the texture of frozen vegetables. Research will continue with green beans, be extended to carrots, and ultimately to other popular frozen vegetables.

8. Microbial Flora in Fruits and Vegetables. Fundamental studies on the microbial flora within tissues of fruits and vegetables have been concluded in Israel, where they were supported by a Public Law 480 research grant to

the Agricultural Research Station of the Ministry of Agriculture at Rehovot. This study demonstrated certain non-pathogenic bacteria in healthy fruit and vegetable tissue. Only certain bacteria are within the tissue. Many organisms found on the surface are not found inside unless the fruit has been damaged; the flora found in fresh fruit remain harmless and have only limited growth in the intact plant. The organisms do multiply during certain processing operations such as brining where bacterial growth is necessary to the fermentation involved in preservation. In this case conditions must be arranged so that the fermenting bacteria will overcome the harmful bacteria present within the fruit and vegetables, or undesirable bacterial growth may bloat or putrefy the brined product.

B. New and Improved Food Products and Processing Technology

1. Foam-mat Drying. Laboratory and pilot plant investigations continue on the application of the foam-mat drying process invented by Department engineers at Albany, California. Informal cooperation continues with industry representatives who are interested in the commercial application of foam-mat drying. Three commercial-scale foam-mat drying units now exist. Two have already been used for commercial-scale research, and the third has produced commercial lots of foam-mat dried tomato paste, pure lemon juice, and lime concentrate. A procedure was developed to further reduce the moisture content of already foam-mat dried powders to eliminate the necessity for in-package desiccants for extremely hygroscopic materials. Rotating vacuum drying equipment, using lime as a moisture acceptor and an electrically heated outer cylinder was designed. Orange powder, an extremely hygroscopic material, was reduced from 3-1/2 to 1% moisture by tumbling 2-1/2 to 3 hours at 130° F.

A method was developed to reduce the bulk and improve the apparent color of foam-mat dried powders. The powders were run between hot rolls to increase their density and provide for more efficient packaging. Conditions for rolling various products were established based on the sensitivity of the products to temperature and the requirements of density specifications. For example, foam-mat dried orange powder was rolled at temperatures between 130 and 180° F. while tomato powder was rolled at temperatures as high as 205° F. The bulk density of foam-mat dried tomato powder has been approximately doubled by this process to give a value of about 0.7 grams per milliliter. The rate of reconstitution in cold water is not significantly changed.

Foam-mat dried tomato powder from concentrated tomato juice was more stable than a commercial preparation of tomato paste. In order to achieve maximum yield of tomato solids in paste manufacture, more severe heat treatment and finishing operations are involved than tomato juice. Seed, peel, stem, and fiber fragments find their way into tomato paste. These constituents may be responsible for poor stability of tomato powder made from commercial paste. Investigations are continuing to determine what components and processes may be involved in this reduction of tomato powder stability.

2. Dry Bean Products. The slow cooking of dry beans and peas deters their utilization. The preparation of dry bean, pea, and lentil powders by drum drying slurries of cooked legumes with and without seasoning was previously reported. Samples of these products have been widely demonstrated with the assistance of the manager of the California Lima Bean Advisory Board. Basic research and research on other bean products is partly supported by the California Lima Bean Advisory Board. Packaging requirements and storage stability of bean powders were studied. Satisfactory flavor was obtained with nitrogen-packed products at 5% moisture stored at 100° F. for six months. Air-packed samples and powders at 10% moisture content were much less stable. An antioxidant in the air-packed sample improved stability but not as much as nitrogen packing. Investigations continued to develop processes which would reduce the cooking time for dry Lima beans. Beans were treated with solutions of food grade substances capable of: (a) complexing metallic ions; (b) dissociating protein complexes and solubilizing proteins; and (c) retarding oxidative processes. Beans treated with these chemicals and dried were completely hydrolyzed and tenderized in boiling water within 30 minutes. The cooked beans had acceptable appearance, texture, and flavor. Shear press evaluations of the freshly prepared beans indicated that the texture of the treated beans was comparable with beans soaked overnight in the usual way and cooked 40 minutes or longer.

3. Processing Quality of Vegetable Varieties. The evaluation of processing quality of vegetable varieties is a continuing program in cooperation with the Washington State Experiment Station at Puyallup, Washington. Increasing attention is being given in western Washington to the introduction of new processing vegetables. With improved varieties of Brussels sprouts available and new developments in mechanical harvesting, larger packs of Brussels sprouts and lower cost to the consumer will be possible. Almost all of the domestic pack of frozen Brussels sprouts is grown in a small coastal area south of San Francisco in California. It appears likely that other growing areas, and western Washington in particular, have conditions for commercial production of Brussels sprouts if improvements in variety and harvest methods can be developed. Interest in western Washington is also focused on the Romano bean and the Fava bean. Varieties of these two vegetables have been grown experimentally at the Western Washington Experiment Station and frozen by Department scientists for evaluation. In addition, 23 varieties of peas and 20 varieties and selections of snap beans were frozen and evaluated in cooperative studies with horticulturists.

4. Frozen and Dehydrofrozen Vegetables. Analyses and evaluations of stored frozen and dehydrofrozen peas indicate little or no difference in color stability between the two products under rather severe storage conditions (i.e., 45 days at 20° F.). There has long been a question as to whether the concentration of reactants resulting from the dehydrofreezing process would harm stability. However, this does not seem to be the case and the dehydrofrozen peas have as good color stability as frozen peas. The color of frozen Brussels sprouts has always been a commercial problem. Brussels sprouts

are so large and so variable in size that uniform blanching is difficult. Overblanching results in loss of their bright green, garden-fresh color. Blanching studies were conducted over a broad range of conditions. Analyses of chlorophyll conversion are completed and results of these studies are currently being evaluated and will be published as a guide to commercial freezers of this vegetable.

5. Fouling of Heat Transfer Surfaces. In the concentration of fruit juices and tomato products, fouling of heat transfer surfaces causes serious economic losses. In earlier work, a resistance-impedance thermometer system was developed to obtain accurate surface temperatures and related heat transfer measurements that were used to discover that fouling was markedly more rapid when products were being warmed compared to when they were being boiled, and to indicate that fouling was increased by increasing surface temperature, vapor fraction in evaporator tubes, and product viscosity. A relationship between protein denaturation and fouling has led to the initiation of studies of the effect of proteolytic enzymes on evaporator surface fouling. Measurements are being extended to rotating steam coil evaporators as they gain commercial acceptance. Simpler generalizations are being sought so that information from these studies can be applied to existing evaporator installations as well as to the design of new evaporators.

6. Microbiology of Frozen Vegetables. In recent years an increasing awareness of microbial sanitation has come to the vegetable freezing industry. Large institutional buyers of frozen vegetables that will be used for remanufacture have specified maximum viable count of bacteria as a quality requirement. Department scientists, through informal cooperation with the vegetable freezing industry in the Pacific Northwest, have conducted plant surveys to assist in improving sanitation procedures. Microbial surveys were carried out during the last year on 5 pea, 1 bean, and 5 corn lines. A typical source of serious contamination was frequently found in improperly sanitized conveyor belts. In one case a 50- to 60-fold increase in bacterial count in peas was observed following passage over two short inspection belts. Contamination from these belts was reduced to a negligible level by continuous spraying with chlorinated water. Use of chlorinated water in fluming in vegetable processing plants has materially reduced bacterial contamination. Such informal cooperation will continue by arrangement out of the Puyallup field station in the State of Washington.

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Area 9 CASTOR, SAFFLOWER, AND OTHER WESTERN OILSEEDS--PROCESSING AND PRODUCTS

Problem. To provide valuable diversification crops for the acreage withdrawn from the production of cotton, wheat, feed grains, and other surplus crops, we must expand the markets for crops such as castor and safflower. But these crops are so new to our agricultural economy that their market potential has not been adequately developed. Castor and safflower have good potential because of the unusual properties of their oils. The possibility of large-scale increases in the production of these oilseeds would be strengthened if high-quality feed products could be developed from the oilseed meals. Basic information is needed on the composition of the oils and of the meals left after extraction of the oil, and this, in turn, requires the development of adequate analytical methodology. Rapid and accurate analytical methods are needed to control and improve the processing of the oils and meals for food, feed and industrial applications. Research on chemical conversion of the oils and evaluation of the modified products is needed to find new or improved large-volume uses. The high percentage of linoleic acid (essential fatty acid) in safflower oil points to a rapidly expanding use as a food oil. But this same fatty acid imparts a high susceptibility to autoxidation. Research is needed to stabilize safflower oil in various food products. Improved procedures for decorticating and processing castor and safflower seeds are needed. There is a particularly critical need to remove or destroy the allergenic and toxic components of castor meal which presently limit its use to fertilizer. Research to isolate and characterize the constituents in castor and safflower meals is needed to develop non-toxic, non-allergenic feed and food products of high value. Basic and applied research is needed to prepare chemically modified products from the meals for industrial applications, to develop economical procedures for carrying out the modifications, and to evaluate the modified products.

USDA PROGRAM

In the Western Utilization Research and Development Division, both basic and applied research are conducted on castor seed at the Division headquarters at Albany, California and, under contract, at Tucson, Arizona. Basic, compositional studies on castor seed meal are concerned with the resolution of its water-soluble proteins and determination of the allergenic and antigenic properties of these components. Studies are conducted on the composition of castor oil, and new analytical techniques are developed.

Applied research on castor meal has as its objective the development of economical methods for deallergenizing the meal without impairing its nutritive quality, to increase its value as an animal feed ingredient. Castor oil and its major constituent, ricinoleic acid, are being studied

to provide for them new and improved industrial applications. Thus, methods are being developed for the preparation of various types of polyurethane foams incorporating castor oil or its derivatives. Procedures are also being devised for the preparation of chemical derivatives of ricinoleic acid, including a number of amides and phosphate esters. Several of the latter compounds may be useful for improving the flame-resistance of castor-based polyurethane foams of the type which may be used for building insulation. The utility of various polymerizable monomers from castor oil for the production of synthetic polymers for use in rubbers, plastics, etc., is being investigated under contract.

The Federal program of research in this area totals 12.2 professional man-years, including contract research equivalent to approximately 0.3 professional man-years per year. Of this total, 2.8 are assigned to chemical composition and physical properties; and 9.4 to new and improved products and processing technologies.

REPORT OF PROGRESS FOR USDA AND COOPERATIVE PROGRAMS

A. Chemical Composition and Physical Properties

1. Detection of Allergens. A risk-free and highly specific test for human allergy, with far-reaching implications for future medical practice and research, was developed in connection with research on allergy induced by castor seed proteins. Through the use of monkeys, the cost of allergy testing has been materially reduced and, more important, the hazards to human volunteers have been eliminated.

Informal cooperation has been maintained with Dr. Raphael Panzani of Marseille, France, who has been supported in part by the American castor oil industry. Reaginic sera of 26 castor-allergic patients from Marseille were tested, confirming the existence of multiple allergies to contaminants of castor pomace dust as well as to components of castor itself. Extensive tests with the sera from Dr. Panzani's allergic patients have unequivocally confirmed the value of the test procedure using monkeys. This new test for allergy provides the basis for solving problems caused by allergen sensitivity to castor and other agricultural products. In addition it promises to be a useful research technique not only in the study of all types of allergy, but also in other fields of medicine and biochemistry.

2. Allergenic Proteins. Chromatographic and electrophoretic separations of castor seed proteins and test for allergenicity indicated that castor seed proteins contain at least six specific antigenic compounds. Not all who are sensitive to castor meal are sensitive to the same antigen in the meal. Conclusive tests were conducted to show that cross-reactivity is involved. Castor pollen and blossoms can cause allergic reactions to pomace-sensitive persons. Furthermore, cross reactions were demonstrated with other members of the spurge family of plants. Contamination of castor

seeds with foreign materials is increased by mechanical harvesting, and contaminants were shown to complicate the allergy problem of castor seed pomace. It is now apparent that insects, molds, and weeds can cause allergic reactions and these foreign materials included in the oilseed pomace may promote sensitivity in humans working with it.

Some of the antigens isolated from castor seed meal have low molecular weights, probably less than 3,000. Low molecular weight antigens may be very potent allergens because the light molecules are easily inhaled into the deep recesses of the lungs, and hence easily absorbed into the blood. The wide distribution of low-molecular weight antigens through the blood stream to all the reactive organs involved in anaphylactic shock may account for the serious and sometimes fatal reaction in sensitized individuals. The existence of low-molecular weight antigens handicaps any deallergenation process involving hydrolytic cleavage of proteins. A successful process must degrade antigenic proteins to polypeptides of lower molecular weight than those occurring naturally in castor meal and capable of functioning as allergens.

B. New and Improved Products and Processing Technology

1. Chemical Derivatives. The ease of preparation of hydroxy-fatty acid amides in high yields and purity was reported previously and has already attracted industrial interest. One of the large castor oil companies is now making the diamide of ricinoleic acid and ethylene diamine as a direct extension of Department pioneering work on the preparation and characterization of a series of amides derivable from ricinoleic acid. The new amide synthesis has been extended to additional fatty hydroxy acids and applied to selected amino acid derivatives. Higher yields and increased optical purity have been achieved. Quarter-pound samples of several amide derivatives of ricinoleic acid have been made and are being evaluated for industrial application. Samples of vinyl-12-hydroxy-stearate, a derivative of castor oil, have been supplied for contract work on the development of polymers at the University of Arizona in Tucson.

2. Urethane Foams. Because of its high content of hydroxy-unsaturated fatty acid, castor oil and castor oil derivatives are useful in the preparation of urethane foams. The lowest cost urethanes can now be produced by employing a combination of castor oil and hydroxyl-containing amines. This formulation for urethane foams is based on Department research. Such foams have a very high insulating capacity and are particularly useful in construction where low thermal conductivity, high strength, and fire resistance are needed.

Flame resistant castor-based rigid foams were prepared using several commercially available fire retardants. Foams prepared with chlorinated castor oil had improved flame resistance and compressive properties as well. Fire retardants containing hydroxy groups react with the isocyanate of the urethane formulation and become part of the polymer when solvent-

blown, rigid urethane foams are prepared. Samples containing 10 to 15% of such retardants, produced self-extinguishing foams with compressive strength and other properties equivalent to controls. Non-reactive retardants produced foams with considerable loss of compressive strength.

The effect of aging on thermal conductivity of foams was investigated. After two months' aging, the desirable low thermal conductivity was retained in polyurethane samples based on both castor oil and polyethers if the polymer skin remained intact. Thermal conductivity increased somewhat if cut foams were aged for two months.

3. Animal Feed Meal. Deallergenized castor meal would constitute a suitable ingredient for livestock and poultry feeds and as such would command a higher price than it does as a general purpose agricultural fertilizer. Several chemical treatments which show promise for a deallergenating castor meal have been studied. Dilute ammonium hydroxide appears the most promising thus far studied. Such treatment is relatively mild, part of the ammonia may be recovered and reused, and the ammonia remaining in the pomace should benefit ruminant feeds.

On a very small-scale laboratory test, the allergens in wet castor seed pomace have been inactivated by ammonia at elevated pressures and temperatures. Non-allergenic pomaces have been tested with guinea pigs, monkeys, and castor-sensitive humans. Small amounts of isolated proteins from castor seed have been added to aliquots of extracts of such pomace. Amounts of allergens too low to cause reaction in humans could be detected in monkeys. With the cooperation of Dr. Panzani in Marseille, 60 of his patients were tested using the non-allergenic pomace and pomace that had been refortified with protein. It appears that correlations between human, monkey, and guinea pig reactions can be used to ascertain maximum safe levels for allergenicity. Such testing could be used to establish the safety of treated castor pomace.

Large-scale digestions of castor pomace have been attempted in a steam-jacketed digestion chamber using gaseous ammonia at 62 lbs. per square inch pressure and 145° C. Fifteen- to thirty-minute treatments gave considerable reduction of allergenicity. However, with 30 minutes of such severe treatment, a hardened, black, refractory material was produced. Further studies will be conducted to seek conditions on large scale that reproduce those of the small laboratory experiments in which successful deallergenation of castor pomace was achieved.

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Area 10 SUGAR BEETS-- PROCESSING AND PRODUCTS

Problem. Sugar beets are mainly processed for sugar; a very small proportion is used for livestock feed. Sugar beets are declining in sugar content and rising in impurities. The traditional processing methods for sugar manufacture cannot cope with beets whose lower quality is due in part to excess nitrogen fertilizer, used to improve tonnage yields. Improved processing procedures should benefit both the growers and processors. It is known that small concentrations of certain chemicals in beets affect processing quality but not enough information is yet available to devise new economical procedures for high-impurity beets. Because costs of producing beets and processing sugar are rising whereas consumption and price of sugar are essentially constant, all factors important to utilizing the crop must be examined to improve processes. There is still too much ignorance of the composition of sugar beets, juices, pulp, and crude sugar to achieve this objective. Sugar losses resulting from spoilage and respiration of beets held at processing plants cannot be prevented by existing methods. Only an expanded research program can provide the needed information at an early date.

USDA PROGRAM

Both basic research and process development studies on sugar beets are being conducted in the Western Utilization Research and Development Division's headquarters laboratory at Albany, California and under P.L. 480 grants, in Calcutta, India, Jerusalem, Israel, and London, England. The basic research program involves a comprehensive study of the naturally-occurring sugar beet and beet juice constituents, both carbohydrate and non-carbohydrate. Biochemical studies of the carbohydrate constituents aim to determine their origin leading to the development of methods for the reduction or elimination of those which decrease sucrose. Chemical studies of the non-carbohydrate constituents provide information to ameliorate effects of adverse constituents. Although Federal research on new products from sucrose (sucrochemicals) has been terminated, studies continue under P.L. 480 grants on the reactions of sucrose with vegetable and animal fats and oils, to produce new and useful compounds having special hydrophilic and lipophilic properties. Processing research on sugar beets deals with the effects of the many variables which influence the efficiency of recovery of sucrose. Pilot-scale sugar beet processing facilities are used to test these processing variables and to evaluate new and improved processing techniques.

The Federal program of research in this area totals 5.8 professional man-years. Of this total, 2.1 are assigned to chemical composition and physical properties; 3.7 to new and improved products and processing technology. In addition the Division sponsors, under P.L. 480, one basic research project and two on product developments.

REPORT OF PROGRESS FOR USDA AND COOPERATIVE PROGRAMS

A. Chemical Composition and Physical Properties

1. Carbohydrate Constituents. Basic studies are under way to isolate and purify non-sucrose carbohydrates from beets for use in studying reactions that interfere with sucrose recovery. A one-step methylation procedure was adapted to convert beet carbohydrates into methylated derivatives volatile and stable enough for separation by gas-liquid chromatography and thin-layer chromatography so they could be identified. Previous studies indicate that galactinol, a beet carbohydrate, as well as raffinose, interferes with the yield of sucrose and should be further investigated. Pure galactinol has now been crystallized and is available as a reference standard in studies aimed at improving the efficiency of sugar recovery from beets.

Investigations on galactose and galactose-containing constituents of sugar beets led to a simple, sensitive test for galactose. This test is now helping to diagnose a rare but severe metabolic disease in infants. This disease, called galactosemia, occurs in certain children born unable to metabolize galactose, and gives rise to jaundice, liver damage, cataracts, and mental deficiency. It can be successfully treated only when detected within a few days after birth. Treatment is to avoid galactose in the diet. The test can be used routinely to diagnose galactosemia by detection of galactose in urine or blood samples and to check foods for galactose so that a safe diet can be prescribed. An enzyme, galactose oxidase, required for this test is now commercially available.

2. Non-Carbohydrate Constituents. The role that chlorides play in the recovery of sucrose from beet juice has been previously reported. When the content of chloride is high, sucrose recovery is low. The recovery of sugar from beets, the chloride content of molasses, and the purity of molasses were found to be closely related. Now it has been shown that the chloride content of thin or thick juice, as well as the chloride content of molasses, is useful in predicting the purity of the molasses that will be produced. Preliminary tests of juice from two factories, using a chemical method to determine sucrose remaining in the molasses, agreed with the purity value predicted by the chloride analysis.

Contract research on the effect of genetic and agronomic factors on non-sugar constituents of sugar beets has been concluded at Colorado State University. The final report indicated that a new F_1 hybrid sugar beet variety has lower chloride and soluble nitrogen contents under all fertility levels studied than the control beets. High yielding beet varieties with low chloride and nitrogen impurities despite heavy fertilization would materially increase returns to growers, while eliminating troublesome and costly processing problems.

3. Enzymic Sucrose Degradation in Sugar Beets. A study of the enzymic sucrose degradation in sugar beet tissues, to provide information necessary for development of improved procedures for handling beets for processing, was initiated under P.L. 480 at the Hebrew University, Jerusalem, Israel. Experimental procedures were developed to handle the beet roots after harvest, to extract beet tissue to provide maximal enzymic activity, and to adapt various quantitative analytical and chromatographic procedures. Several intermediate chemical compounds, especially in the nucleotide-sugar category, were synthesized with radioactive carbon or phosphorus as substrates in the enzymic studies. Several enzymic pathways are being studied to obtain more information on the biosynthesis of sucrose in the beet root.

B. New and Improved Products and Processing Technology

1. Sucrochemicals. The domestic Federal program of research on sucrochemicals was terminated in response to recommendations of the Sugar Research and Marketing Advisory Committee. Prior to termination, negotiations had taken place to provide P.L. 480 grants for work in this area. At the Tropical Products Institute in London, England, novel and patentable processes were discovered for preparing from sucrose and derivatives of animal fats compounds exhibiting a high degree of surface activity which may be of considerable commercial value as surfactants. In addition to providing a market for sugar, they could increase use of animal fats which are presently in critical surplus. Chemical processes were discovered to provide greatly simplified, direct routes for preparing ethers of organic polyhydroxy compounds. With a highly reactive molecule, sucrose, serving as the polyhydroxy compound, long-chain alcohol derivatives from animal fats are readily combined to form ethers. Such ethers, for example, mono- and di-dodecyloxymethyl ethers of sucrose have considerable surface activity and may also find use in wetting, dispersing, penetrating, and emulsifying agents. A patent application is being filed in the United States to cover the processes discovered and, when issued, will be assigned to the United States Government and be available for free licensing by manufacturers within this country.

Basic research on the reaction of sucrose with sulfonyl chloride and similar compounds, to provide substances for manufacture of polymers, was initiated at Jadavpur University, Calcutta, India. Initiation was hampered by difficulty in obtaining trained scientists and import licenses for essential chemicals and apparatus. However, a literature search was made and a review was published to form a firm foundation for the laboratory work that is just being initiated.

2. Juice Diffusion. Investigations are continuing with the objective of improving diffusion processes whereby a higher recovery of sugar from beets may be obtained in the initial extraction. It was previously reported that laboratory and pilot plant tests indicated an improved method of utilizing the two usual sources of water for sugar diffusion (i.e., well water

normally containing natural salts, and distilled water recovered from the evaporation of sugar juices). By adding the stream of well water at the tail end of the diffuser and the pure distilled water somewhere toward the head end, purer sugar juices were obtained. The impurities in the well water were largely eliminated by the exhausted pulp as it leaves the diffuser. One beet sugar company, operating in an area where well waters are saline, has applied the diffusion scheme with good success and others are currently evaluating the process.

Another investigation towards improving the diffusion of sugar from beets is concerned with an alkaline treatment of the cut beets to modify the natural pectin. About 30% of sugar beet insoluble solids is pectin. Alkali will interact with the pectin in sugar beet cossettes. At low temperature the principal reaction breaks chemical linkages in the large pectin molecule forming pectic acid and acetic acid. At high temperatures the large pectin molecule is broken into smaller fragments by degradation. Degradation reduces the mechanical strength of the tissue and hinders diffusion of sugar and dewatering of exhausted pulp. Laboratory trials indicated that a carefully controlled lime treatment of sugar beet cossettes at low temperature stabilizes the pectin molecule and enhances the diffusion, leaving a tough, readily pressed pulp. More water can be pressed from the pulp providing a greater quantity of thin juice and a drier, more easily handled pulp; the overall lime consumption is less than in usual diffusion-plus-carbonation of the juice to remove impurities at a later stage in the refining. Variables resulting from lime treating the cossettes will be subjected to further laboratory examination and pilot plant studies prior to cooperative trials on larger scale equipment.

3. Juice Purification. Laboratory tests were conducted to find better methods for purifying thin juice extracted from sugar beets. Preliminary work with static columns indicate that glucose and ionic impurities can be separated from sucrose by ion and molecular exclusion. Removal of such impurities, if it can be accomplished on a commercial scale, should improve the efficiency of sugar recovery from beets. Experiments made with a hot column using diluted molasses showed better purification of sucrose and higher concentration than similar experiments using a cold column. However, there appear to be one or more amino compounds that cannot be easily separated from sucrose using hot columns, even though removal of color in certain fractions was very good. Investigations will continue to seek methods for improvement in purification methods.

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New and Improved Products and Processing Technology

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1/ Research supported by P.L. 480 funds.

Area 11 POULTRY-- PROCESSING AND PRODUCTS

Problem. The $1.6 billion poultry meat industry operates on very narrow profit margins. This industry is confronted with the problem of converting continually increasing amounts of poultry into a wide variety of products having high quality and improved convenience, at costs attractive to consumers and remunerative to the poultry grower. Information on the properties and processing of poultry is not sufficient to enable us to better utilize poultry in a variety of forms attractive to consumers. Increased utilization of poultry would also serve toward eliminating our feed grain surplus, increasing returns to farmers and providing better products for American consumers.

Although poultry is an efficient converter of feed to meat, more grain is used by poultry per calorie of food produced than by any other commercial animal because a high percentage of the poultry diet is grain and because poultry meat contains exceedingly little fat. Furthermore, one-fourth of all grain fed to animals is used for poultry and egg production. Hence, increased consumption of poultry products would be an effective means of increasing markets for surplus grain. Also, the efficiency of feed utilization by poultry makes possible low prices within reach of more consumers. A still further benefit would arise from the increased use of poultry by improving the nutrition of consumers having diets now low in animal protein.

The consumption of poultry has steadily increased from a 1947-1949 average of 22 lbs. per capita to 37 lbs. for 1962. This important increase has involved price, quality of product, availability, and disposable income. Because of the current low profit margin it is impractical to increase consumption by lowering farm prices. Increased demand for and consumption of poultry will require higher quality and more convenient products and a greater variety to meet the desires of the modern consumer. However, in addition to greater returns from increased demand, a greater profit margin for the farmer can, of course, come from greater efficiencies in processing.

The trend toward convenience foods and further processing has primarily led to precooked poultry products which are generally less stable, more subject to warmed-over flavors, and more likely to provide texture problems than uncooked items. With the expansion of operation and the emphasis on continuous, more efficient processing, need has arisen for improved processing procedures for feather removal, chilling, tenderization, freezing, deboning, and commercial cooking. Lowering the cost and improving the quality of products that can be stored at ambient temperatures, such as canned, dried, cured, and irradiated products, offer potential for poultry utilization in domestic and export markets. As a foundation for applied studies, further knowledge is needed on the chemical nature of flavor and flavor changes in processing and storage, on tenderness development, and on proteins, lipids, and other components.

USDA PROGRAM

Basic and applied research on poultry meat and poultry meat products are conducted at the Division headquarters at Albany, California and, by contract, in East Lansing, Michigan. Fundamental studies on poultry flavor are concerned with the identification of flavor precursor constituents in poultry meat and in the isolation and identification of volatile flavor components developed during the cooking of poultry. The chemistry of muscle protein and post-mortem chemical changes are investigated relative to the tenderness and other quality characteristics of poultry. The basic physiological character of feather release mechanism in fowls is studied to provide a foundation for improved feather removal. Applied research is conducted on the stability of cold-tolerant organisms; special problems of flavor, texture and stability of precooked frozen foods; and processing factors that influence tenderness of poultry meat.

The Federal program of research in this area totals 20.2 professional man-years, including contract research equivalent to approximately 0.9 professional man-years per year. Of this number, 9.4 are assigned to chemical composition and physical properties; 10.8 to new and improved food products and processing technology.

REPORT OF PROGRESS FOR USDA AND COOPERATIVE PROGRAMS

A. Chemical Composition and Physical Properties

1. Flavor Chemistry. In basic studies on the flavor of poultry meat, emphasis is placed on the isolation and identification of constituents of the volatile fraction of poultry that has been cooked by various procedures. The object of these studies is to establish the relation between natural flavors or off-flavors, and these constituents, particularly those that occur in precooked, frozen, and canned poultry. Studies of the volatiles from fried chicken were completed. Results confirmed indications that off-flavor in processed chicken comes from an increase of natural components rather than a development of new components. This suggests that quality of poultry flavor is in large part a function of concentration of volatiles. For one volatile, hydrogen sulfide, it was shown that muscle proteins are the principal precursors. Non-proteins such as glutathione, taurine, methionine, and cystine contribute little to the total hydrogen sulfide released when poultry is cooked.

Tenderizing procedures, including aging of slaughtered chicken and antemortem adrenalin injection, were observed to affect chicken broth flavor. The treatments cause a small difference in acidity. However, in other tests, a pH adjustment of 0.4 pH unit upward or downward after cooking and a pH difference as great as 0.17 pH unit during cooking did not affect chicken broth flavor. Therefore, the flavor differences between tenderized and untreated chicken cannot be attributed to pH differences.

2. Post-Mortem Biochemistry and Tenderness. Basic investigations were conducted on the component proteins of poultry muscle and their reactions post-mortem which affect tenderness and other texture qualities. The pattern of tenderization in poultry is as follows: meat cooked a few minutes after slaughter is more tender than meat allowed to age for one hour before cooking. The transient period of initial tenderness before onset of rigor mortis, is difficult to demonstrate in poultry because the very act of cooking accelerates the metabolic processes in the meat. Rapidity of onset of rigor in young birds was not found necessarily to result in toughness. Chemical changes accompanying rigor mortis include breakdown of glycogen and adenosine triphosphate and increase in acidity. Glycogen breakdown was closely associated with the development of toughness. With the normal occurrence of glycogen breakdown, the meat rapidly becomes tough and remains tough until the aging process, in some unknown manner, renders the meat tender again. Acceleration of post-mortem glycogen breakdown in young poultry increases the toughness of fully aged meat. Injections of epinephrine or of iodoacetate prior to slaughter or very rapid cooking were tried to eliminate or reduce post-mortem glycogen breakdown. Meat from birds in which post-mortem glycogen breakdown was eliminated or inhibited is initially tender and remains so through a 24-hour aging period.

3. Physiology of Feather Release. The physiological mechanisms that control feather tightening and release in poultry are being investigated in contract studies at Michigan State University at East Lansing. The involvement of the nervous system in the mechanisms by which physical and chemical agents influence feather release, was studied. The direct involvement of the central nervous system was demonstrated by performing spinal section. The feather pulling force in the dorsal feather tract, posterior to the section, was markedly reduced, while there was little effect anterior to the section. When the disconnected end of the severed cord was stimulated electrically, the feather pulling force returned to its high normal value during the stimulus. A functional relationship between a nerve located just below the skin surface and the feather pulling force in the femoral feather tract was established in an anesthetized bird. The nerve that influenced feather release was traced back to the lumbar-sacral plexus. The failure of injected curare to alter plucking indicates that the skeletal voluntary muscles do not take part in the release of feathers.

B. New and Improved Food Products and Processing Technology

1. Low-Temperature Microbiology. Fundamental and applied research was conducted to determine growth, survival, and lethal conditions for microorganisms that grow at temperatures of 32° F. and lower. A literature survey on low-temperature microbiology was completed, a valuable convenience to other scientists, and a sound foundation for Department research. Freezing storage and repeated freezing and thawing of chickens were found to have no adverse effects on subsequent keeping time under refrigeration. Salt brine dips prior to freezing or refrigeration increased keeping time as much as 25%. Similar treatment in water containing 50 p.p.m. of free chlorine

increased keeping time by the same amount. Preliminary studies indicated that proper incubation temperature for bacterial plates used for bacterial analysis of chilled foods should be at a temperature close to that at which the food was stored, rather than at 35° or 37° C. as is commonly used. Long-term storage experiments with frozen chicken products inoculated with high levels of bacteria, were initiated to determine whether bacteria in a frozen product where they are not growing will affect storage stability.

Questions have been raised concerning the bacterial safety of cooked turkey rolls. Bacterial levels in the center of turkey rolls were reduced markedly during cooking. Counts were reduced to an insignificant level at center temperatures of 153° F. Cooking temperatures of 165° and higher are generally recommended for cooking this product.

The Department scientist specializing in low-temperature microbiology is serving on an international and two national committees concerned with the development of microbial standards for food products, thereby providing a first-hand advisory function in this specialized line of investigation.

2. Freeze-Drying of Poultry. A new investigation was initiated on the freeze-drying process which converts foods to a stable, lightweight form which may be easily reconstituted so as to be practically indistinguishable from the original state. Freeze-drying is now a substantial commercial operation. Its full potential can be achieved only by reducing cost, which is high because very good vacuum must be maintained and cycle times are extremely long for food pieces of the usual size. The object of the new work is to reduce processing cost, partly through reducing the drying cycle time, partly through reducing the cost of equipment. The ultimate goal is a cheap, continuous, high-capacity freeze dryer. Two engineering configurations are being explored. In both the mass of frozen pieces is tumbled in vacuum with radiant heat on them to sublimate ice. Basic design features are being explored. Upon completion of this work a prototype dryer will be designed and constructed for use on large experimental scale.

3. Tenderness and Other Textural Qualities. Use of polyphosphates in the water used to chill turkeys prior to freezing benefits frozen cooked meat stored at 10° F. for nine months. In chilling tests with frozen chicken fryers, polyphosphates were found to improve retention of water during refrigerated holding or cooking. Moisture uptake during slush ice chilling was reduced by the use of polyphosphates and spoilage during subsequent holding under refrigeration inhibited. It is presumed that chelation of trace metal compounds by the polyphosphates prevents bacterial growth.

Department work aimed at improving meat tenderness is being coordinated with that of research institutes. Several lines of attack have been taken up against the tenderness problem, including histological studies, studies of water binding and inorganic salt effects, changes in the solubility of protein, the role of enzymes, post-mortem glycolysis, effects of age, breed, sex, and feeding practices, and the influence of ante-mortem and post-mortem

handling practices and cooking methods. Coordination between agencies is one of the functions of the Institute of American Poultry Industries, whose research committee includes Department scientists.

4. Radiation Preservation of Poultry Products. Irradiation-sterilized poultry is considered potentially attractive to feed troops where refrigeration is not available. With funds transferred from the Department of Defense, Quartermaster Research and Engineering Command, evaluation of poultry preserved by several radiation treatments was undertaken. Major problems of flavor, color, and texture losses during processing and subsequent storage were revealed and means for controlling some of the damage were developed, particularly to protect color and flavor. Heat inactivation of enzymes and temperature control during radiation treatment eliminated a red discoloration and reduced off-flavors.

5. Precooked Frozen Poultry Products. It was also shown that starchy cereal thickening agents were effective in reducing the liquid exudation that commonly occurs during baking of prepared turkey meat loaves after freezing and frozen storage.

The freezing and reheating of fried chicken increases the tendency of the usual coating materials to peel off and become unattractive. Partial cooking shrinks the meat before the batter is applied so that the coating adheres to the meat through freezing, thawing, and reheating. Both shrinkage of the meat during cooking and the pressure of escaping moisture tend to loosen coatings. Research demonstrated how batter ingredients can be varied to obtain desired thickness, crispness, and color. Results can be applied by processors to develop a variety of coatings that will provide the adhesive quality, thickness, appearance, and consistency needed to increase the value of fried chicken and other breaded frozen products.

PUBLICATIONS REPORTING RESULTS OF USDA AND COOPERATIVE RESEARCH

Chemical Composition and Physical Properties

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Area 12 EGGS-- PROCESSING AND PRODUCTS

Problem. The $1.8 billion egg industry is periodically faced with burdensome surpluses that drive prices below the break-even point for many producers. The industry is also faced with declining per capita consumption. The estimate for 1963 is down 18% from the 1947-1949 average consumption. Because the demand for table eggs is inelastic, the increased utilization of eggs must come in the form of new egg products that compete by means of quality and convenience. Adequate knowledge is lacking of the properties, processing characteristics, and new product potentials of eggs to develop new markets. Present outlets for the 10% of egg production that is frozen or dried include the baking, confectionery, salad dressing, noodle, and baby food trades. Modified and new products emphasizing quality and convenience are needed to increase acceptance of egg products by these industries in order to compete successfully with egg substitutes.

Increased utilization of eggs would not only benefit the producer, but would also diminish our feed grain surpluses since poultry and egg production account for about one-fourth of all grain fed to animals. Improved egg-containing products would benefit the producer in three ways: by providing an increasingly useful buffer for stabilizing egg prices; by providing additional uses and outlets for eggs; and by providing more remunerative outlets for wholesome eggs that are unsuitable for table use because of appearance or handling characteristics.

Egg processors have four general problems. First, the potential of yolk-containing solids in convenience foods can be fulfilled only with improvement of flavor stability, of dispersibility, and freedom from pathogenic Salmonella bacteria. Secondly, the processing costs of whites must be reduced and their utility improved in order to dispose of whites which accumulate in surplus because of the demand for yolks. Third, further basic research on egg composition and components is essential to reach an understanding of physical and chemical changes induced by processing and storage and thus provide a rational basis for devising improved processes and products. Fourth, formulation studies designed to incorporate eggs into new household and institutional convenience products, are needed. This last study must encompass a full appraisal of physical, chemical, and microbiological problems peculiar to the formulated products.

USDA PROGRAM

In the Western Utilization Research and Development Division, a broad program of basic and applied research is conducted at the Division headquarters at Albany, California; by contract in Austin, Minnesota, and Ames Iowa; and by grant funds under P.L. 480 in France and India. Fundamental research is

conducted on egg proteins and their relations to the functional properties and quality of eggs, on egg lipids and their role in off-flavor development in yolk solids, on the mechanism of bacterial penetration and survival in eggs, and on the bactericidal, antiseptic, anti-inflammatory, and food preservative properties of lysozymes and other components from eggs. Applied research is conducted on the stabilization of yolk-containing solids to increase the usefulness of eggs in dry mixes and other convenience foods, on new and improved drying procedures to make dried egg fractions and products more readily and more completely dispersible, on various methods of controlling Salmonella in eggs, and on factors in the handling of shell eggs that affect egg product quality and cost.

The Federal program of research in this area totals 18.6 professional man-years, including contract research equivalent to approximately 0.9 professional man-years per year. Of this number, 6.3 are assigned to chemical composition and physical properties, 12.3 to new and improved food products and processing technology. In addition, two research grants on basic problems are supported by P.L. 480 funds.

REPORT OF PROGRESS FOR USDA AND COOPERATIVE PROGRAMS

A. Chemical Composition and Physical Properties

1. Egg Proteins. Basic investigations on lysozymes from various sources, including eggs, are conducted at the University of Paris, France, supported by P.L. 480 funds. Lysozymes are enzymes with bactericidal and anti-inflammatory properties. The amino acid composition of egg lysozymes has been compared with lysozymes from a number of other animal sources, including poultry. Lysozymes were found to vary in biological activity; those having greater activity contain more basic amino acids. Substrates for the enzymic action of lysozyme are being isolated and analyzed as a step towards the elucidation of the relationship between chemical structure and biological activity of lysozymes. Lysozymes were purified from the egg white of duck's and hen's eggs. Comparative studies are under way using lysozymes from bacterial and other animal sources.

2. Bacterial Spoilage of Shell Eggs. Basic research on spoilage of shell eggs is continuing. Dilution techniques used in the laboratory counts of various Pseudomonas species which are involved in egg souring, were modified using peptone water as a diluent, in order to eliminate the killing effect of various other dilution media. The killing effect of dilution media in bacteriological population counts was connected with minor traces of copper. Removal of this heavy metal by redistillation, ion exchange, or treatment with hydrogen sulfide followed by filtration eliminated the destructive effect. Pseudomonas species were found to be especially susceptible to heavy metal contamination, while other food spoilage bacteria are not sensitive to copper in the concentrations found in distilled water from a piping system.

The contribution of metabolic products of egg spoilage bacteria to the bacteria's ability to infect eggs is being investigated. A fluorescent pigment is excreted by the Pseudomonad egg spoilers and the presence of this pigment is used to detect eggs infected and spoiled by these bacteria. This pigment was purified and preparations isolated in suitable quantities for chemical study. At least four separate compounds are present in the fluorescent pigment. All four of the separated compounds bind ferric iron ions. This observation lent support to the hypothesis that iron-binding compounds excreted by spoilage bacteria counteract the antibacterial iron-binding action of conalbumin and other proteins of egg white. The Pseudomonad species from which the pigment had been obtained is unable to grow in egg white, because the small amount of iron required for growth is complexed with the protein, conalbumin and unavailable to the bacteria. However, supplementation of egg white with as little as ten micrograms per milliliter of a crude but iron-free pigment preparation obtained from growing bacteria, was followed by extensive growth of the bacteria. It is now thought that the pigment can recapture iron from the conalbumin complex and hold it in a form that is available to support bacterial growth. These findings explain the mechanism by which Pseudomonads infect and spoil eggs. Infection on the shell and at membranes is a common occurrence. Growth of bacteria at the membranes results in the formation of the pigment which diffuses into the white. The presence of the pigment therein permits extensive multiplication of the bacteria and spoilage of the egg.

3. Oxidative Changes in Yolk Lipids. Basic studies are conducted by contract at the Hormel Institute of the University of Minnesota aimed at elucidation of the oxidative mechanism of egg lipid systems to serve as a basis for interpreting and correcting undesirable oxidative flavor deteriorations induced by processing and storage of eggs. Lecithin prepared from fresh eggs exerted an antioxidant effect when added to egg powders to be stored. In contrast when lecithin was prepared from egg powders that had been stored it had a pro-oxidant effect on the autoxidation of methyl linoleate. Preliminary studies indicate that carotenoids in eggs also exert a pro-oxidant action on the egg lipid system. An improved laboratory technique to recover volatiles from stored egg powders has been developed to measure oxidative changes as egg products are stored.

B. New and Improved Products and Processing Technology

1. Egg Powders. Dispersion and rehydration were materially advanced by gas-impregnation spray drying. A gas impregnating system compatible with continuous spray drying on high-capacity equipment was developed and successfully applied to egg yolk using carbon dioxide, nitrous oxide, or gaseous nitrogen. Modifications permit wide variation in particle sizes and bulk densities of products. Particles over 1000 microns in diameter and powders having a bulk density of less than 0.1 gram per ml. have been readily obtained. Free-flowing, whole egg powders of improved dispersibility were found to perform better in layer cakes and cookies than those agglomerated after drying or dried at atmospheric pressure after being whipped into a foam.

2. Control of Salmonella in Egg Products. Three aspects of research and development are involved in Salmonella control in processed egg products. First is research toward more reliable analytical methods that can be used to determine whether or not viable Salmonella exist in any product; secondly, a study of the bacteriological factors, including degree of contamination and conditions of pasteurization necessary to kill organisms; and finally an investigation of changes in flavor and functionality of egg products for baking and other uses, that are caused by heat treatments to control the bacteria.

Improved heat treatments and equipment are sought to kill Salmonella in eggs under conditions which do not damage the eggs. Steam infusion and conventional plate heating and holding tube equipment were both used to pasteurize liquid egg. Application of direct steam infusion to flash heat whole egg to 160° F. for less than 5 seconds, followed by vacuum flash cooling to 125° F. destroyed inoculated Salmonella of average heat resistance in whole egg at about 1000 viable organisms per gram. This heat treatment did not cause significant baking function loss in either frozen or spray dried products. Steam infusion heating of egg liquid appears promising as a means of reducing bacterial contamination of egg products. However, demonstrations are still to be conducted on commercial-scale equipment under normal processing conditions.

Dried egg white is stable at ambient temperature only if reducing sugars have been removed prior to dehydration. One method of removing reducing sugars is to ferment liquid egg white with bacteria. Such fermentations, when controlled, limited the development of Salmonella in egg products. An alternate method for minimizing the reducing sugar-induced protein deterioration and discoloration also inhibits Salmonella. Addition of table sugar or other suitable carbohydrate and adjusting the pH of the liquid egg white improved storage stability and prevented discoloration of stored dried egg white, but slightly diminished functional properties. This method, like fermentation desugaring would be of use where freedom from Salmonella was more important than maximum performance quality.

4. Precooked Frozen Egg Products. Principles governing the behavior of essential basic ingredients in foods that influence their suitability for use in prepared and precooked frozen foods are under investigation. Special emphasis is placed on the foaming, emulsifying, thickening, binding, and gelation properties of eggs and cereals since products having structures involving these properties are particularly susceptible to freezing damage. The relation of solids content to stability of foam products, such as soufflés, is being investigated. Soufflés with high solids maintain their volume through freezing and reheating, although soufflés with lower solids had a greater volume than those with higher solids content immediately after baking. However, their volume diminished on reheating after they had been stored at 0° and 10° F.

PUBLICATIONS REPORTING RESULTS OF USDA AND COOPERATIVE RESEARCH

Chemical Composition and Physical Properties

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1/ Research supported by P.L. 480 funds.

Area 13 PHARMACOLOGY

Problem. Advances in agricultural science and processing technology necessitate the use of an increasing number of new chemical compounds whose safety must be established. The mutual interests of both the producers and the consumers of agricultural products, demand that public agencies participate in securing unequivocal evidence of safety before products of advanced technology are marketed. This responsibility is particularly acute where a public agency, such as the Department of Agriculture, contributes to technological developments that result in intentional or unintentional addition of untested components into foods, feeds, or into materials contacting the persons of consumers, or developments that result in the introduction, concentration, or modification of natural components in a way that may have an adverse physiological effect on consumers. Types of materials that require continuing surveillance include food additives, inadvertent residues of pesticides and other useful agricultural chemicals, antibiotics and medicinals, and the naturally occurring chemical constituents of physiological importance. In the areas of interest to Agricultural Utilization Research and, in particular, in connection with process and product developments of the four Utilization Research Divisions, such compounds must be tested by short- and long-term ingestion in experimental animals, such as rats and dogs, to secure toxicological data required by the Federal Food and Drug Administration to establish safety and legal certification for their use. The unequivocal establishment of safety for any useful chemical involves much more than merely conducting animal feeding tests on a routine service basis. It often requires original chemical analytical procedures and metabolic fate studies in experimental animals, as well as new methodology and observational techniques, to study of new chemicals. Each assignment in this field is a new area for original, often fundamental research.

USDA PROGRAM

Pharmacological investigations supporting the Department's utilization research and development program are conducted in the Western Utilization Research and Development Division at Albany, California. Agricultural products, and additives required to preserve or otherwise treat them, are investigated as they may cause toxic or allergenic reactions. Laboratory methods for discovering the metabolic fate of chemical compounds in animal physiology are developed and applied to problems in the utilization of farm products. Plant constituents that exert deleterious or beneficial effects on animal growth are studied to determine quantitative responses.

The Federal program of research in this area totals 10.0 professional man-years assigned to pharmacology investigations. Additional pharmacological studies directed toward specific commodities are reported elsewhere and include 2.8 professional man-years assigned to chemical composition, physical and physiological properties of castor, safflower, and other western oilseeds

(reported in Area No. 9 of A Summary of Current Program, 7/1/63; and Preliminary Report of Progress for 7/1/62 to 6/30/63.

REPORT OF PROGRESS FOR USDA AND COOPERATIVE PROGRAMS

A. Pharmacology Investigations

1. Cottonseed Components. Acute oral toxicity of 15 cottonseed pigment gland fractions prepared at the Southern Regional Research Laboratory is being investigated. Preliminary LD_{50} values were obtained as a guide for further fractionation work aimed at isolating toxic factors other than gossypol. In 15 fractions the LD_{50} acute toxicity ranged from 1700 mg/kilo of body weight to 5000. On the basis of these results all of the pigment gland fractions tested would be considered slightly toxic or practically non-toxic. This suggests that acute toxicity data have less practical significance for evaluating the toxicity of fractions of pigment glands than data for continued ingestion over a period of time. Cottonseed meal was compared with soybean meal at 30% levels in the diet of weanling male and female rats for 90 days, followed by breeding tests. Neither diet had an adverse effect on growth or breeding.

Feeding tests with male and female weanling albino rats for 90 days at dietary levels of 15% were conducted using three oils: (1) a cottonseed oil containing cyclopropene, (2) a cottonseed oil from which cyclopropene had been removed by refining and bleaching, and (3) corn oil. Organ weights were determined but revealed no significant differences. Samples of body fat and liver were sent to the Southern Regional Research Laboratory for stearic and oleic acid analyses. The only abnormality encountered to date was a slight growth inhibition in the female rats ingesting the two cottonseed oils. Reproduction studies were conducted and five out of six rats on the cyclopropene-positive cottonseed oil cast litters. The results are extremely good in comparison with control colony breeding.

2. Toxicity Tests--Replacement Crops. The agricultural value of several plants which are under investigation as potential sources of industrial oils, is dependent upon a by-product use of seed meal, generally for livestock feed. Feeding tests were conducted on Crambe abyssinica whole seed, hulls, meal, and fractions to determine toxicity. Several other Crambe samples in short supply were also assayed. Most samples have been found to be inferior to the control basal diet in terms of growth rate and efficiency of food utilization. A Crambe meal fraction prepared at the Northern Regional Research Laboratory and representing 3/4 of the original meal gave improved growth, whereas the protein and thioglucoside fractions removed were found to inhibit growth. Growth of rats on a diet containing 30% Veronia anthelmintica marc was inferior to a comparable group on 30% soybean meal. Approximately half of the inhibition was corrected by addition of methionine to the diet. Parsley seed meal at a 20% level in the diet produced the same growth rate as an equal amount of soybean meal. Growth on Limnanthes douglasii meal was only slightly reduced relative to soybean meal. Addition of methionine had no effect.

Dimorphotheca meal was decidedly inferior to soybean meal and there was no improvement obtained by addition of methionine and lysine. Toasting Dimorphotheca meal to eliminate heat-labile factors was ineffective. Cassia bonariensis and Cassia occidentalis seeds were quite toxic at dietary levels of 5%.

3. Soybean Meal Investigations. Growth inhibition is observed when raw soybean meal is fed to rats. Steam heating of soybean meal at atmospheric pressure for as little as 15 minutes will eliminate the growth inhibition and pancreas-stimulating factor. Increasing the heating time to 2 hours had no further beneficial or detrimental effects. Tempeh (a mold-fermented soybean product) has been fed to rats with results indicating that feed value tends to be reduced for short periods of fermentation rather than enhanced by such treatment. Although isolated soybean protein in purified diets contains a factor which reduces the availability of dietary zinc, data were obtained to indicate that the growth inhibition when raw soybean meal was fed is not due to lack of zinc in the diet.

4. Research on Forages. Biological activity of components of alfalfa and clover is assayed with laboratory animals. Pharmacological tests are made of concentrates, extracts, and fractions of these products. Much attention has been devoted to coumestrol from alfalfa, which exhibits estrogenic activity in mice. Several new samples of alfalfa containing coumestrol at levels of 200 to 500 p.p.m. make it possible to measure the activity of the alfalfa meal rather than an extract therefrom. Unexpectedly, when these alfalfa meals were fed to mice, the estrogenic response calculated from chemical assays was not realized. However, if the acetone extract of the meal, which also contains coumestrol, is fed, then a three-fold increase in uterine weight was observed, indicating estrogenic activity. The possibility exists that an estrogen inhibitor may be a natural component of the alfalfa meal. An observation from an agricultural research laboratory in Australia is also pertinent. The isoflavone formononetin has been found to exhibit considerable estrogenic potency to sheep. This plant estrogen, when assayed in mice showed low biological activity. A re-evaluation of the mouse assay as a criterion of potency in ruminants must be initiated and may be of great importance to investigations of plant estrogens in forage.

5. Dry Beans. The composition of dry beans is being investigated to determine the factor or factors involved in their intestinal effects. Animal studies are continuing at Albany, California, both by feeding beans to rats and by injecting bean components in their upper intestines. As previously reported, irritation of intestine walls and intestinal swelling have been observed. Research procedures have been refined so that sections of the intestine can be excised and weighed. It has been found that weights of treated strips of intestines are 50 to 70% heavier than controls when an injection of ethanol extract of beans has been made into an intestinal loop. Earlier tests were made with pentobarbital anesthesia which restricts peristalsis. Recently urethane that allows peristalsis to continue has been substituted. The injection of the alcoholic fraction with urethane results

in increased peristalsis and distension of the intestine with gas and with mucus. The largest volume of gas was produced when bean extracts were inserted into the rat's stomach by stomach tube. Starved rats were used for this experiment and peptone was added to stimulate the flow of gastric juices. Striking demonstrations of the effect of an alcoholic extract of beans were obtained using this method.

6. Caloric Availability Studies. Caloric availability of several materials was evaluated by an assay procedure involving the measurement of body weight gains of young rats receiving the test material as a supplement to a restricted caloric intake. Invert sugar was used as a caloric standard. Raw honey was available to rats at a rate of 4.04 calories per gram. Bulgur wheat wafers formulated in connection with development of a Civil Defense shelter ration were 91% digestible by rats and contained 4.04 calories per gram. Several altered fats were assayed, providing the following digestibility values: amylose palmitate #23, 31%; amylose palmitate #34, 13%; amylose stearate, 21%; succino-stearin, 41%; Bis(2,3 dihydroxypropyl) adipate, 88%; and Bis/1-(hydroxymethyl)-2-(stearoyloxy)ethyl/ adipate, zero digestibility. Limnanthes seed oil, petroselinic acid, and capric acid were comparable to corn oil as energy sources. Parsley seed oil was not readily ingested by rats and was poorly utilized. Both high and low molecular weight dextrans were well absorbed and utilized. For comparative purposes agar and pectin were tested and found to be poorly digested (20% digestibility) and not utilized. It was impossible to evaluate the caloric availability of a diet with 10% Vernonia oil because the rats would not eat it.

7. Toxic Fescue. All attempts to produce in experimental laboratory animals symptoms that would reproduce or explain the toxic responses in cattle ingesting toxic fescue have failed to date. These tests involved the feeding of high dietary levels of fescue to rats, mice, and guinea pigs; the intravenous injection, subcutaneous injection, and administration by stomach tube of extracts of fescue. The injection of fluid extracts of fescue into roosters has failed to produce a cyanosis of the comb as in the case with ergot alkaloid. Tests on the smooth muscle of intestine and uterus with alcoholic extracts of fescue by the Schultz-Dale technique have consistently demonstrated the presence of a histamine-like substance which causes a contraction that is antagonized by adrenaline. However, this reaction is non-specific and is produced by extracts of non-toxic fescue, ladino clover, alfalfa, peanuts, and castor meal. A crystalline alkaloid isolated from toxic fescue failed to contract smooth muscle with the Schultz-Dale technique.

8. Polysaccharide B-1459. The polysaccharide gum B-1459 was fed to male and female beagle dogs at about 10 and 20 grams per dog per day, approximately equivalent to 1 and 2 grams per kilo of body weight per day. At the end of 30 days there were no adverse effects other than some weight loss on both dosage levels and diarrhea on the high dose.

9. α-Sulfo Fatty Acids. Sodium salts of α-sulfo fatty acids are surface active and may have potential value as detergents and wetting agents. Twenty-two α-sulfo fatty acids were tested for acute toxicity by administration by stomach tube into mice. All compounds were relatively non-toxic.

Area 14 REPLACEMENT CROPS-- UTILIZATION POTENTIAL

Problem. Farmers could achieve more economic use of their land if new and profitable crops were available that would have new end-use patterns. For example, it would be advantageous to develop a new oilseed crop yielding fatty acids that could find industrial use in applications for which acids from presently available domestic oilseed crops are unsuitable. To develop a new crop, three basic steps are involved: (1) survey of wild plants, in cooperation with plant scientists, to identify those having both potentially valuable components and promising agronomic potential; (2) detailed physical and chemical characterization of components of interest to obtain clues to likely end uses; (3) selection of the most promising species, followed by additional research to explore uses and demonstrate industrial potential, and by additional agronomic research to establish proper cultural practices and select the best strains and varieties. Only after these steps have been successfully accomplished can a proposed new crop be offered to agriculture and industry for introduction and development. Obviously, a program of this type is a long-range one. Yet, whether the future of agriculture involves conditions of surplus, of greater emphasis on foods and feeds, or of pressure for greater national self-sufficiency, the nation will benefit from availability of practical crop plants to serve its needs.

To achieve the objective, survey and characterization work needs to be greatly increased, since the greater the number of species examined, the greater will be the opportunities for finding plants meeting the criteria of high utilization and agronomic potential. Work of the Department has already revealed several promising sources of new potentially valuable water-soluble gums, pulp fibers, and oils containing unique fatty acids such as hydroxy-unsaturated acids, capric acid, epoxidized acids, and unusual long-chain fatty acids. In order to demonstrate the potential of these new materials, further work is required on their physical and chemical properties and reactions, on processing to obtain maximum recovery from source plants, and on by-products from processing, such as oilseed meals.

USDA PROGRAM

Basic and applied research is being conducted on hydroxy-unsaturated acid-containing oilseeds, in the Western Utilization Research and Development Division's headquarters laboratory at Albany, California; and by contract at Fargo, North Dakota. The basic, compositional studies emphasize the development of special analytical techniques for application to new oils containing hydroxy-unsaturated fatty acids. In the applied area, research is conducted to develop and evaluate industrial products from the hydroxy-unsaturated oils.

The Federal program of research in this area totals 4.0 professional man-years, including contract research at a rate equivalent to approximately 0.2 professional man-years per year. Of this total, 2.0 are assigned to chemical composition and physical properties; and 2.0 to industrial utilization.

REPORT OF PROGRESS FOR USDA AND COOPERATIVE PROGRAMS

A. Chemical Composition and Physical Properties

1. Dimorphotheca and Lesquerella Seed Oils. Seed of the species Dimorphotheca sinuata are rich in an oil that contains dimorphocolic acid. The fatty acid composition of Dimorphotheca oil has been determined with isolations accomplished using partition, gas-liquid, and thin-layer chromatography and identifications involving ultraviolet, infrared, and nuclear magnetic resonance spectroscopy. About 2/3 of the fatty acid content of this oil is dimorphocolic acid, about 1/8 linoleic, and about 1/10 oleic acid. The presence of less than 1% of an epoxy acid has been definitely established by nuclear magnetic resonance analysis, and a hitherto unrecognized component, 9-keto,10,12-octadecadienoic acid, has been found in the amount of almost 2-1/2%.

Seeds of wild members of the mustard family, the Lesquerellas, are a source of two new fatty acids. Lesquerolic and densipolic acids have been found in two Lesquerella species and appear to have potential industrial uses. Lesquerella oils bear some similarity to castor oil, and are expected to have industrial uses in coatings, plastics, and chemical intermediates. Derivatives of Lesquerella oil have been made by high temperature pyrolysis of methyl lesquerolate in a procedure similar to that used to prepare derivatives of ricinoleic acid from castor oil. Good conversions of methyl lesquerolate to heptaldehyde and methyl tridecylenate have been obtained. Secondary decomposition products have been obtained also in small proportions. Conversion of the glycerol esters of fatty acids to methyl esters has been made substantially quantitative by using a high ratio of methanol-to-oil in a continuous system employing strongly basic ion exchange resins as catalysts.

These new derivatives of Lesquerella and Dimorphotheca oil are adding to the wealth of substances to be screened for industrial utilization.

B. Industrial Utilization

1. Industrial Products from Hydroxy-Unsaturated Oils. Continued preparation of new derivatives from Lesquerella and Dimorphotheca seed oils is beginning to provide novel compounds for industrial evaluation. Vinyl-9-hydroxy-stearate prepared from Dimorphotheca oil has been supplied for evaluation in making polymers at the University of Arizona in a contract research project. Long-chain phosphite esters of hydroxy fatty acids are being prepared for evaluation as plasticizers. A contract investigation has been initiated at North Dakota State University to evaluate the use of Lesquerella and Dimorphotheca oil derivatives in protective coatings.

Line Project Check List -- Reporting Period July 1, 1962 to June 30, 1963

Work & Line Project Number	Work and Line Project Titles	Work Locations During Past Year	Line Proj. Incl. in Summary of Progress	Area & Sub-Subheading
W1 2-24 (Rev.)	Air classified flours	Albany, Calif.	Yes	1-A-1 1-B-2
W1 2-27 (Rev.)	Wheat feed products	Albany, Calif.	Yes	1-C-1
W1 2-29 (Rev.)	Bread flavors	Albany, Calif.	Yes	1-A-8
W1 2-31	Effects of processing on properties and new product development	Albany, Calif.	Yes	2-A-1 2-B-1 2-B-2
W1 2-36[1]	Lipoglutenin studies	Albany, Calif.	Yes	1-A-7
W1 2-38[1]	Parboiled wheat foods	Albany, Calif.	Yes	1-B-1
W1 2-39 (C)[1]	Effect of additives on bread flavor	Cambridge, Mass.	Yes	1-A-8
W1 2-40 (C)[1]	Estrogens in wheat germ and bran	Ames, Iowa	Yes	1-C-1
W1 2-41	Wheat endosperm constituents	Albany, Calif.	Yes	1-A-3
W1 2-42 (C)	Methods for determining wheat proteins	Pullman, Wash.	Yes	1-A-3
W1 2-43 (Rev.)	Gluten foods	Albany, Calif.	Yes	1-B-2
W1 2-44	Protein interactions	Albany, Calif.	Yes	1-A-1
W1 2-45 (C)	Water-dispersible protein preparations	Lafayette, Ind.	Yes	1-B-2
W1 2-46 (C)	Chemical basis for cohesiveness in gluten	Kansas City, Missouri	Yes	1-A-4
W1 2-47 (C)	Elimination of microbial contaminants of wheat flour	Chicago, Ill.	Yes	1-B-6
W1 2-48 (C)	Identification of wheat proteins by radiotracer techniques	Pullman, Wash.	Yes	1-A-1
W1 2-49 (C)	Protein and lipid composition of spring and winter wheat	Manhattan, Kansas	Yes	1-A-6
W1 2-50	Mechanism of flour maturation	Albany, Calif.	Yes	1-A-6
W1 2-51	Compositional factors of wheat relative to continuous-mix processes	Albany, Calif.	Yes	1-B-4
W1 3-16 (Rev.2)	Improved forage feed products	Albany, Calif.	Yes	3-B-2
W1 3-18	Phenolic components of forages	Albany, Calif.	Yes	3-A-1 3-A-2 3-B-1 3-B-2
W1 3-19 (C)	Autoxidation of alfalfa lipids	Berkeley, Calif.	Yes	3-A-3
W2 2-7 (Rev.2)	Molecular properties of wool and mohair proteins	Albany, Calif.	Yes	4-A-1
W2 2-11 (Rev.)	Minimizing degradation of wool by acids and alkali	Albany, Calif.	Yes	4-A-1
W2 2-22 (Rev.)	Chemical treatment of wool for shrink resistance and other "easy-care" properties	Albany, Calif.	Yes	4-B-1 4-B-4
W2 2-24	Effect of fabric construction and functional properties	Albany, Calif.	Yes	4-B-3
W2 2-26[1]	Setting and relaxation of fibers in wool fabrics	Albany, Calif.	Yes	4-B-5
W2 2-28 (Rev.)	Mechanical behavior of wool fibers and fibrous assemblages	Albany, Calif.	Yes	4-A-2
W2 2-29 (Rev.)	Effects of radiation on natural and modified wools	Albany, Calif.	Yes	4-A-3
W2 2-30	Nuclear magnetic resonance absorption of natural and modified wool and mohair	Albany, Calif.	Yes	4-A-1
W2 2-32	New types of yarns and fabrics from coarse wools	Albany, Calif.	Yes	4-B-1

[1] Project discontinued during the reporting period.

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Work & Line Project Number	Work and Line Project Titles	Work Locations During Past Year	Line Proj. Incl. in Summary of Progress	Line Proj. Incl. in Area & Sub-Subheading
W2 2-33 (C)	Improved bleaching of wool	Lowell, Mass.	Yes	4-B-6
W2 2-34 (C)	Chemical modification of wool to increase drying rate	Durham, North Carolina	Yes	4-A-1
W2 2-35 (C)	Wear-wrinkling performance of light weight wool fabrics	Washington, D. C.	Yes	4-A-2
W2 2-36	WURLAN treatment of wool top	Albany, Calif.	Yes	4-B-1
W3 1-67 (Rev.2)	Processing quality of Northwest soft fruit and berry varieties	Prosser and Puyallup, Wash.	Yes	6-B-3
W3 1-83 (Rev.)	Flavonoids in citrus	Pasadena, Calif.	Yes	5-A-2 5-A-6 5-B-1
W3 1-88 (Rev.)	Citrus essential oils	Pasadena, Calif.	Yes	5-A-1 5-B-1
W3 1-95 (Rev.)	Composition of dates and date products	Pasadena, Calif.	Yes	5-A-4 5-B-2
W3 1-101 (Rev.)	Fruit juice products and processes	Albany, Calif.	Yes	6-B-4
W3 1-109 (Rev.)	Processing quality of Northwest grapes for juice	Prosser, Wash.	Yes	6-B-8
W3 1-112 (Rev.)	Dried fruit products and processes	Albany, Calif.	Yes	6-A-2 6-A-6 6-B-2 6-B-5
W3 1-113[1]	Tree nuts products and processes	Pasadena, Calif.	Yes	6-B-7
W3 1-116[1]	The chemistry of sulfur dioxide in dried fruits	Albany, Calif.	Yes	6-A-5 6-B-2
W3 1-117 (Rev.)	Fruit pigments	Albany, Calif.	Yes	6-A-1
W3 1-119	Fruit flavor components	Albany, Calif.	Yes	5-A-3 5-B-4 6-A-4 6-B-1
W3 1-120 (C)	Macadamia nuts products and processes	Honolulu, Hawaii	Yes	6-B-7
W3 1-121	Heat transfer surface fouling	Albany, Calif.	Yes	6-B-9 8-B-5
W3 1-122	Texture of fruits and fruit products	Albany, Calif.	Yes	6-B-6
W3 1-124 (C)	Cell wall organization of fruits	Cambridge, Mass.	Yes	6-A-3
W3 1-125	Composition of desert grapefruit	Pasadena, Calif.	Yes	5-A-5
W3 1-126	Viniferous grape products	Albany, Calif.	Yes	6-B-8
W3 1-127	New fruit dehydration methods	Albany, Calif.	Yes	6-B-5
W3 4-47 (Rev.)	Vegetable varieties	Prosser and Puyallup, Wash.	Yes	8-B-3
W3 4-74 (Rev.)	Composition of dry beans re processing factors and product quality	Albany and Pasadena, Calif.	Yes	8-A-3 8-B-2
W3 4-75	Measurement and preservation of chlorophyll in vegetables	Albany, Calif.	Yes	8-B-4
W3 4-77 (Rev.)	Tomato concentrate and powder	Albany, Calif.	Yes	8-A-5 8-B-4
W3 4-79 (Rev.)	Effects of processing on potato product flavor	Albany, Calif.	Yes	7-A-1 7-B-1
W3 4-80	Effects of processing operations upon texture of frozen vegetables	Albany, Calif.	Yes	8-A-7
W3 4-81 (C)	Dry bean characteristics	Urbana, Ill.	Yes	8-A-3
W3 4-82	Microbiology of frozen vegetables	Puyallup, Wash.	Yes	8-B-6
W3 4-83	Mechanism of anhydration in bacterial spores	Albany, Calif.	Yes	8-A-2
W3 4-84 (C)	Histological studies of vegetables for dehydration	Davis, Calif.	No [2]	

[1] Project discontinued during the reporting period.

[2] Recently initiated project.

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Work & Line Project Number	Work and Line Project Titles	Work Locations During Past Year	Line Proj. Incl. in Summary of Progress	Area & Sub-Subheading
W3 4-85	Flavor of tomato products	Albany, Calif.	Yes	8-A-1
W3 4-86	Chemistry and enzymology of vegetable flavors	Albany, Calif.	Yes	8-A-1
W4 3-1	Chemical derivatives of ricinoleic acid	Albany, Calif.	Yes	9-B-1
W4 3-2 (Rev.)	Foamed polyurethanes from castor oil	Albany, Calif.	Yes	9-B-2
W4 3-3 (Rev.)	Pharmacology of castor bean allergens	Albany, Calif.	Yes	9-A-1 9-A-2 9-B-3
W4 3-5 (C)	Polymerization of castor oil-derived monomers	Tucson, Ariz.	Yes	9-B-1
W4 3-6	Role of blossoms and pollen in castor allergy	Albany, Calif.	Yes	9-A-2
W4 3-7 (C)	Characterization of antigenic proteins of castor	Menlo Park, Calif.	No [3]	
W4 3-8	Castor pomace deallergenation	Albany, Calif.	Yes	9-B-3
W5 1-72 (C)	Relation of beet composition to processing characteristics	Fort Collins, Colorado	Yes	10-A-2
W5 1-73	Biochemical studies of non-sucrose carbohydrates in sugar beets	Albany, Calif.	Yes	10-A-1
W5 1-75	Effects of non-sugar chemicals on processing	Albany, Calif.	Yes	10-A-1 10-A-2 10-B-2 10-B-3
W5 5-37	Evaluation of hydroxy-conjugated dienoic acid oils	Albany, Calif.	Yes	14-A-1 14-B-1
W5 5-46 (C)	Preparation and evaluation of surface coatings	Fargo, North Dakota	Yes	14-B-1
W6 1-41 (Rev.)	Improvement of egg white products	Albany, Calif.	Yes	12-B-1 12-B-2
W6 1-48 (Rev.)	Chemistry of poultry flavor	Albany, Calif.	Yes	11-A-1
W6 1-49 (Rev.)	Microbiology of cold-tolerant organisms	Albany, Calif.	Yes	11-B-1
W6 1-53	Processing characteristics of eggs	Albany, Calif.	Yes	12-A-2
W6 1-54	Precooked frozen foods	Albany, Calif.	Yes	11-B-5 12-B-3
W6 1-55	Improvement of yolk-containing egg solids	Albany, Calif.	Yes	12-B-1
W6 1-56	Tenderness and other textural qualities of poultry meat	Albany, Calif.	Yes	11-A-2 11-B-3
W6 1-57 (C)	Oxidative changes in yolk lipids	Austin, Minn.	Yes	12-A-3
W6 1-58 (C)	Control of the neuromuscular retention and release of feathers	East Lansing, Mich.	Yes	11-A-3
W6 1-59 (C)	Reduction of Salmonella contamination in egg products	Ames, Iowa	No [2]	
W6 1-60 (C)	Histological study of frozen poultry	Madison, Wisc.	No [2]	
W6 1-61	Elimination of Salmonella in egg products	Albany, Calif.	Yes	12-B-2
W6 1-62	Freeze-dried poultry meat	Albany, Calif.	Yes	11-B-2
WU-P-1	Plant enzymes	Albany, Calif.	Yes	6-A-3 6-A-8
WU-0-0-1 (BF)	Hop oil flavor components	Albany, Calif.	Yes	8-A-1
WU-0-0-2	Fallout shelter foods	Albany, Calif.	Yes	1-B-3
QMC-0-11	Radiation preservation of poultry products	Albany, Calif.	Yes	11-B-4
UR-A10-(10)-22	Rheology of wheat flour doughs	Haifa, Israel	Yes	1-A-2
UR-E9-(10)-2	Composition of whole wheat lipids	Paris, France	Yes	1-A-7

[2] Recently initiated project.

[3] Investigation has been terminated without completion of the contract. Final negotiations are pending.

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Work & Line Project Number	Work and Line Project Titles	Work Locations During Past Year	Line Proj. Incl. in Summary of Progress	Line Proj. Incl. in Area & Sub-Subheading
UR-E9-(10)-7	Immunochemical analysis of wheat and barley proteins	Paris, France	Yes	1-A-3
UR-E9-(10)-8	Solubility of wheat gluten proteins	Montpellier, France	Yes	1-A-3
UR-E9-(10)-43	Phosphorus in wheat flour	Paris, France	Yes	1-A-7
UR-E9-(10)-44	Ultrasonic study of wheat gluten	Paris, France	Yes	1-A-3
UR-E9-(10)-45	Enzyme action in low-moisture grain	Paris, France	Yes	1-A-5
UR-E15-(10)-31	Wheat germ proteins	Bologna, Italy	Yes	1-A-3
UR-E21-(10)-1	Sulfhydryl groups in wheat	Poznan, Poland	Yes	1-A-5
UR-E21-(10)-18	Coenzyme role of riboflavin of wheat endosperm	Poznan, Poland	Yes	1-A-5
UR-E29 (10)-14	Wheat flour lipids	Chorleywood, England	Yes	1-B-4
UR-E29-(10)-38	Separation of the total protein of wheat flour	St. Albans, England	No [2]	
UR-E29-(10)-47	Biological value of processed wheat	Cambridge, England	Yes	1-B-5
UR-E29-(10)-52	Structure of alfalfa polysaccharides	Edinburgh, Scotland	Yes	3-A-4
UR-E8-(20)-10	Finishing treatments for improved qualities in wool fabrics	Helsinki, Finland	Yes	4-B-7
UR-E9-(20)-1	Sequence of amino acids in wool proteins as related to quality differences	Lille, France	Yes	4-A-1
UR-E29-(20)-10[1]	Identification of sulfur-containing compounds in wool	Leeds, England	Yes	4-A-1
UR-E29-(20)-11	Penetration of charged molecules into keratins	Leeds, England	Yes	4-A-1
UR-E29-(20)-22	Lubrication of wool knitting yarns	Nottingham, England	Yes	4-B-2
UR-A10-(30)-32	Enzymatic browning in deciduous fruits	Jerusalem, Israel	Yes	6-A-2
UR-E15-(30)-11[1]	Canned concentrated peach and apricot purees	Parma, Italy	Yes	6-B-10
UR-A10-(30)-3[1]	Microbial flora in fruits and vegetables	Rehovot, Israel	Yes	6-A-7 8-A-9
UR-E26-(30)-5	Autoxidation of fats in dehydrated vegetables	Gothenburg, Sweden	No [2]	
UR-E29-(30)-16	Enzymatic browning of potato	Cambridge, England	Yes	7-A-2
UR-E29-(30)-17	Sulfur dioxide in dehydrated vegetables	London, England	Yes	7-A-3 8-A-6
UR-E29-(30)-20	Carotenoid components of vegetables	Cambridge, England	Yes	8-A-5
UR-E29-(30)-27	Relationship of composition to cooking quality of dry peas	Chipping-Campden, England	Yes	8-A-4
UR-A7-(50)-31	Reaction of sucrose with sulfonyl chloride and other chemicals	Calcutta, India	Yes	10-B-1
UR-A10-(50)-25	Enzymatic sucrose degradation in sugar beet tissues	Jerusalem, Israel	Yes	10-A-3
UR-E29-(50)-33	Fatty acid esters of sucrose	London, England	Yes	10-B-1
UR-A7-(60)-27	Physico-chemical properties of hen egg yolk proteins caused by freezing	Bangalore, India	No [2]	
UR-E9-(60)-76	Chemistry of egg lysozyme	Paris, France	Yes	12-A-1

1 Project discontinued during the reporting period.

2 Recently initiated project.